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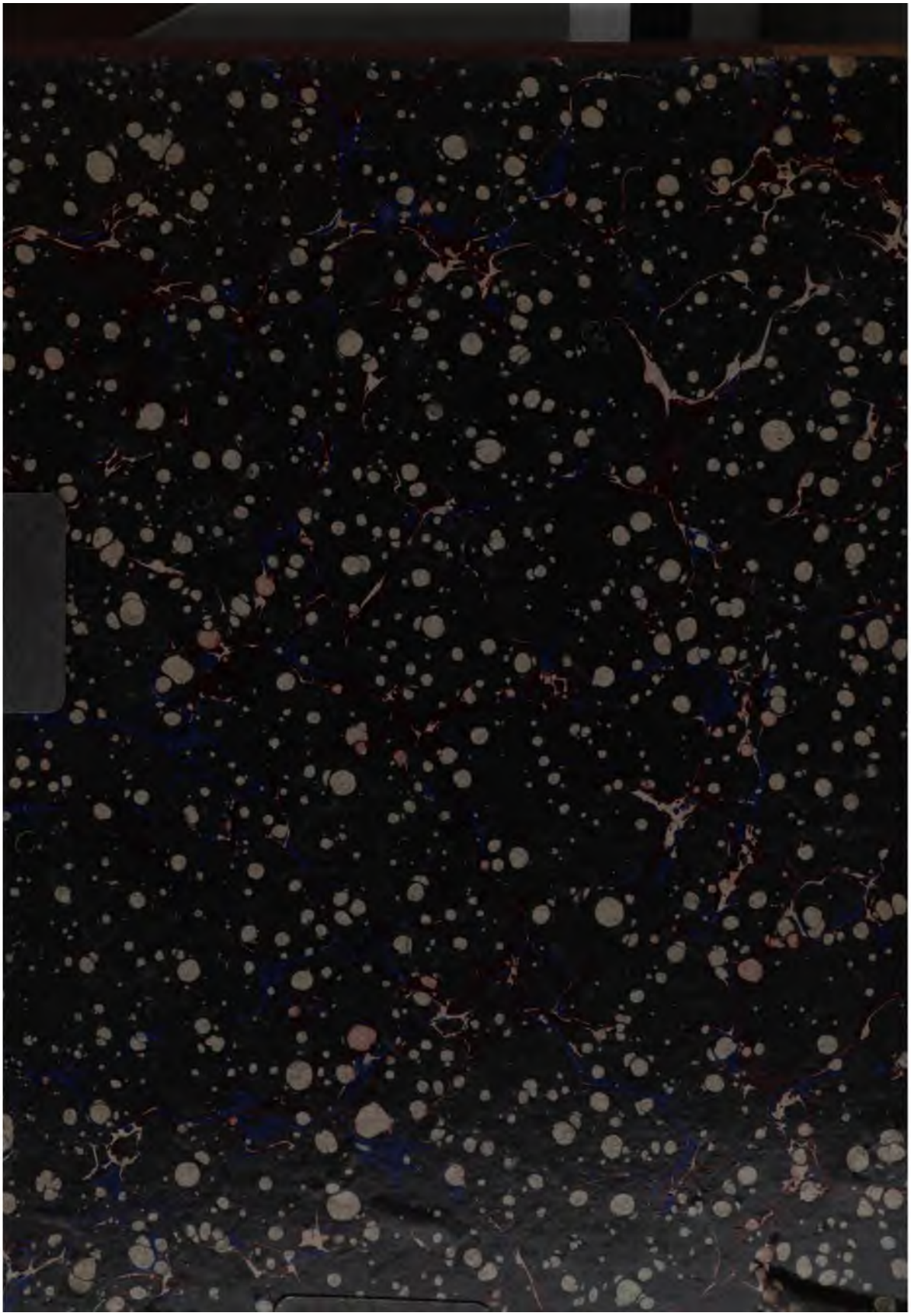

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Description and Distribution

BY

CHARLES PALMER NOTT

WITH NINE PLATES

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April 30, 1904

NITOPHYLLA OF CALIFORNIA. DESCRIPTION AND DISTRIBUTION.[1]

BY CHARLES PALMER NOTT.

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[1] Contributions from the Botanical Laboratories of the University of California, No. 10.

I. INTRODUCTION.

OUR knowledge of the genus *Nitophyllum* Grev. as represented upon the coast of California and adjacent shores has been hitherto of a fragmentary and limited nature. The number of species found in the region named, the points of their occurrence, and their identity with the forms already known, or their title to recognition as distinct species, have been for some time largely matters of conjecture. With a view of providing a more connected account of these species, so far as data could be obtained, the following descriptions and accompanying notes upon distribution are presented.

II. HISTORY.

The first notice of Californian *Nitophylla*, so far as the writer is aware, was made by W. H. Harvey (1858, Pt. II, p. 104, Supp., p. 128), who mentions the species of *Nitophyllum* then known upon the western coast of North America. Two forms, *N. fryeanum* and *N. laceratum (N. violaceum)*[1], were mentioned as occurring at Golden Gate, San Francisco Bay, California. Still later, in a "Notice of a Collection of Algæ made on the Northwest Coast of North America, chiefly at Vancouver's Island, by David Lyall, in the years 1859–61." Harvey (1862, p. 170) gives the original description of *Hymenena latissima (N. latissimum)*, specimens of which were dredged, or found adrift in Esquimault Harbor, Vancouver's Island, B. C.

The next mention of Californian *Nitophylla* is found in the "Bidrag till Florideernes Systematik" of J. G. Agardh (1871, p. 49), where reference is made to Harvey's species *Hymenena latissima* Harv. *(N. latissimum)*.

A noteworthy addition to the number of North American species was made when Professor W. G. Farlow (1875, p. 365) published a "List of the Marine Algæ of the United States, with Notes of New and Imperfectly Known Species."

[1] The names in parentheses are those applied to the species as recognized in this paper, where the quoted name differs.

In this paper were enumerated the following six species: *N. (Neuroglossum) andersonii* J. Ag. *(N. andersonianum)*; *N. fissum* J. Ag. *(N. ruprechtianum)*; *N. fryeanum* Harv.; *N. laceratum* Grev. *(N. violaceum)*; *N. latissimum* J. Ag.; *N. ruprechtianum* J. Ag.

In the following year Farlow (1876, p. 695) published a second list, which included all the above mentioned species with the addition of *N. areolatum* D. C. Eaton *(N. latissimum)* and *N. spectabile* D. C. Eaton.

The most comprehensive account of *Nitophyllum* yet presented was that contained in the Epicrisis Floridearum of J. G. Agardh (1876, pp. 446–472, 698–701). This account of the genus included all of the species then known to occur on the west coast of North America, with the exception of *N. spectabile* D. C. Eaton. To the forms mentioned by Harvey and Farlow were now added *N. multilobum* J. Ag., *N. violaceum* J. Ag., and *N. flabelligerum* J. Ag. *(N. ruprechtianum)*.

Farlow (1877, pp. 238, 245), in a paper discussing some algæ new to the United States, comments on some of the species mentioned by him in the two papers of 1875–76, and also gives the original description of *N. spectabile* D. C. Eaton.

Between 1877 and 1898, the literature pertaining to Californian *Nitophylla* consists of lists of the forms occurring on the coast, with the exception of Hervey's "Sea Mosses" (1881), a popular work, in which some of the characteristics of color, size and venation of the *Nitophylla* are described. Several other writers should here be noticed. Dr. C. L. Anderson (1891, p. 224), M. A. Howe (1893, p. 67), Daniel Cleveland, and A. J. McClatchie (1897, p. 358) have published lists of coast forms. No additions were made to the number of west coast forms by any of these writers except the last named, whose paper contains the first mention of the occurrence of *N. uncinatum* J. Ag. upon the Californian coast.

The latest and most complete statement in regard to west coast *Nitophylla* is found in the volume published by J. G. Agardh (1898).

According to Agardh twelve species of *Nitophyllum* and two species of *Neuroglossum* are assigned to the west coast of North America. The complete list is as follows, viz.: *N. farlowianum* J. Ag. *(N. ruprechtianum)*; *N. flabelligerum* J. Ag. *(N. ruprechtianum)*; *N. fryeanum* Harv. *(N. fryeanum)*; *N. latissimum* J. Ag.; *N. macroglossum* J. Ag. *(N. latissimum)*; *N. multilobum* J. Ag.; *N. marginatum* J. Ag. *(N. ruprechtianum)*; *N. ruprechtianum* J. Ag.; *N. spectabile* Eaton.; *N. stenoglossum* J. Ag. *(N. violaceum)*; *N. uncinatum* J. Ag.; *N. violaceum* J. Ag.; *Neuroglossum andersonianum* J. Ag. *(Nit. andersonianum)*; *Neuroglossum lobuliferum* J. Ag. *(Nit. violaceum ?)*.

Agardh, basing his distinction between species upon differences in color, texture, form of frond, and position of cystocarps, as well as upon the more reliable characters, such as shape and position of sori, and venation of frond, regards all these forms established by him as valid species.

It should be observed, however, in regard to these forms, that the descriptions are in many cases, as admitted by the author himself, drawn up from fragmentary or imperfect specimens. Further, the characteristics which are employed to a considerable extent by Agardh as distinctions between species, and even between subsections, are variable to a marked degree. Observation of a considerable range of forms by the writer has led to the conclusion that sufficient allowance has not been made for the variations which require that great freedom should be used in defining the boundaries of species occurring on the Californian coast. A distinction should be made between the more variable characters such as color, texture, form of frond, and position of the cystocarps, and the less variable characters, such as shape and position of sori, and venation of frond.

After a careful examination of the descriptions of the species as formulated by Agardh, and further study and comparison of the variations of the plants themselves, the writer is obliged to conclude that the degree of elasticity which seems desirable in considering Californian forms has not been permitted in regulating the limits of a species, or

else access has been had to material which has not come under the writer's observation. A further consideration of these forms will be found in the remarks upon species in a later portion of this paper.

The writings of the three authors mentioned above, viz., Harvey, Agardh, and Farlow, constitute the important literature upon Californian *Nitophylla*, and their work alone will be considered in the further discussion of the species.

III. GENERAL DESCRIPTION.

The fuller discussion of the species of *Nitophyllum* of the Californian coast will be advanced by some special treatment of the prominent morphological characters and geographical distribution of the genus itself, for the sake of the increased light thrown by such treatment upon like points in connection with the forms to be hereafter discussed.

The following synopsis exhibits the principal characters of this genus and will be followed by a discussion of some special features of morphology and distribution, as illustrated by Californian species.

SYNOPSIS OF GENERIC CHARACTERS.

Frond either erect or exhibiting a prostrate and an erect portion.

Prostrate frond creeping, linear, or irregularly expanded or membranous, occasionally with midrib, nerves, or veins; with or without rhizoids; variously lobed, divided or branched, occasionally proliferating; branches rising at intervals into erect fronds; margin entire, serrate, dentate, crenate, undulate, or lobed; sometimes forming offshoots and innovations.

Erect fronds rising from holdfast or prostrate frond, singly or several together; sessile, subsessile, or stalked; flat and linear, or membranous, variously lobed, divided, forked, segmented, and branched; frequently proliferating;

with or without midrib, nerves or veins; with margin entire, serrate, dentate, crenate, undulate, or lobed. Branching subdichotomous, subpinnate, subpalmate, or palmate, with branches or segments entire, linear or expanded, sometimes much prolonged. Stalk linear, flat, with or without distinct midrib; frequently becoming thickened and cylindrical through wearing away of margin of frond and renewed growth of remaining portion; frequently twisted by wave action; often persistent and freely proliferating.

Midrib, when present, usually conspicuous, narrow or wide, simple below, sometimes branched above, sometimes evanescent or dividing into flabellate or anastomosing nerves, frequently becoming thickened, stout and persistent, freely proliferating. Nerves usually conspicuous, occupying body of frond, margin, or apices, usually branching freely, flabellate, free, or anastomosing, sometimes dividing into minute and inconspicuous veins. Veins inconspicuous or microscopic, occupying body of frond, margin or apices, simple or branching, flabellate, free, or anastomosing, commonly evanescent and indistinguishable from ordinary tissue of frond.

Sporangia found on both surfaces of the frond, usually in locally thickened portions, in sori of varied shape, containing tripartite tetraspores. Sori minute and scattered over the entire surface of the thallus, or large, forming orbicular patches disposed irregularly over the surface; or linear marginal patches; or lines arranged radially along the margin; or borne on marginal or surface proliferations of varying size.

Antheridia developed from the superficial cells of the thallus, forming whitish patches scattered over the surface of the frond, the latter frequently becoming rugose.

Cystocarps scattered over both surfaces of the frond, or arranged along the margin, or borne on marginal proliferations, usually large, projecting beyond the surface of the thallus, opening by a carpostome.

REMARKS ON SOME GENERIC CHARACTERS, AS ILLUSTRATED BY CALIFORNIAN SPECIES.

The Prostrate Frond.—The prostrate creeping frond possessed by many species of *Nitophyllum* deserves special consideration. Agardh makes some use of this character in separating the genus into subsections, but the importance of this structure to the plant and the extent to which it may be developed have not, so far as it has been possible to learn, been very fully demonstrated.

Influence of Substratum.—The character of the substratum upon which the plant is located and the extent to which it is exposed to the dashing, drawing, or swirling force of the waves affect both the amount of growth and the shape of the prostrate frond.

The various species of *Nitophyllum* occur in a variety of situations, from about mid-tide mark outward and downward to deep water. They grow in some cases upon the piles of wharves, where the prostrate frond must take advantage of cracks in the wood or roughenings of the surface to secure a foothold. Other species are found upon bare rock-surfaces, exposed to the dash of breakers. The most common situation is that of those species which inhabit sheltered rock crevices or pools surrounded by rocks which protect them from the force of the waves. In such spots there will usually be found upon the rocks a rich growth of Bryozoa and Porifera, whose sponge-like substance affords an excellent foothold for the plants and is conducive to a free development of the prostrate frond. . Other algæ, notably the Corallines, afford by reason of their jointed structure and rough surface, excellent habitats. In general, the characters of the prostrate frond correspond to those of the erect portion, but there are marked exceptions. *N. latissimum*, for instance, possesses a membranous, broadly divided, erect frond, while that of *N. andersonianum* is very much branched. Two forms in the same genus could hardly seem more widely different, yet the prostrate frond of the two species is very much alike where growing under

approximately similar conditions of substratum and wave action. Under other conditions the prostrate frond of *N. latissimum* may become widely linear and irregularly lobed, lose the toothed margin which forms one of the points of resemblance to *N. andersonianum*, and acquire a midrib and nerves, in this case resembling the prostrate frond of *N. ruprechtianum*. Again, *N. multilobum*, which as a rule selects sharply inclined or vertical rock-surfaces as a habitat, forms by means of its prostrate frond orbicular patches composed of the closely overlapping, broadly lobed, and membranous portions of the creeping, prostrate frond. The room afforded on the bare rock apparently favors the radial development here exhibited; while the necessity for securing adequate thickness and firmness to meet the dash of the waves has led to the close overlapping or dovetailing of the various divisions of the prostrate frond.

It has already been remarked that the prostrate part of the plant resembles in many respects the erect portion which rises from it. This statement usually holds good with regard to the shape and branching, but does not apply to the venation. As a rule, a midrib or nerves are lacking in the prostrate frond of those species whose erect fronds are provided with such structures.

Formation of Rhizoids.—A number of species also exhibit a response apparently to the stimulus of contact, by sending short processes or rhizoids from the surface of the frond to the substratum to which they adhere. These processes have been observed on the under surface of the prostrate frond of *N. ruprechtianum*, *N. violaceum*, *N. multilobum*, *N. harveyanum* and *N. corallinarum*, and are recorded for several other species. Still more remarkable is the instance observed of the formation of these rhizoids in the case of a plant of *N. violaceum*. This specimen had wrapped itself around portions of the thick frond of *Prionitis lanceolata*, to whose surface numerous processes sent forth from the surface of the *Nitophyllum* in contact with the *Prionitis* had attached themselves.

Offshoots and Innovations.—A point of further interest in connection with the development of the prostrate frond is found in the formation of offshoots and innovations. In the first case, slender branches may arise from the margin of the older portion of the frond. These grow, secure attachment for themselves, and separate from the parent frond, and later give rise in turn to erect fronds. In the second case, by the growth and branching of the prostrate frond, an extended structure is produced, the ramifications of which become separated from each other by the decay and disappearance of the older portions, thus forming innovations in a manner similar to the process occurring in the Bryophytes.

The Erect Frond.—The erect part of the plant commonly rises singly from the prostrate portion, but occasionally the fronds are clustered together, as in *N. multilobum*, where they are grouped in the middle of the orbicular patch formed by the prostrate frond.

Size.—The height and breadth of the erect frond is an extremely variable character. *N. corallinarum*, occurring upon *Corallina chilensis*, does not reach a height of 2 cm.; while *N. spectabile* is reported by D. C. Eaton as reaching 50–60 cm.

Shape and Branching.—Great diversity exists in the shape and branching of the erect frond. Some species are broadly membranous, and but slightly lobed or divided. Good examples of this type are *N. spectabile*, *N. latissimum* and *N. fryeanum*. At the other extreme may be placed such a finely dissected and abundantly branched species as *N. andersonianum*. Between these two opposing types are found all gradations of frond division and arrangement of branches. It is not uncommon to find in one and the same species forms exhibiting a tendency to become broadly membranous, or very much divided and branched. For instance, in *N. ruprechtianum*, there seems to be a tendency toward the flabellate or expanded type of frond, though the typical specimens of the species are characterized by division of the frond into linear, much prolonged

branches. *N. violaceum*, on the other hand, varies in the direction of the finely dissected type of frond. *N. fryeanum* is a membranous form which often becomes narrowly laciniate, with the segments much prolonged. *N. andersonianum*, which has been instanced as an example of a finely dissected frond, becomes, when growing in quiet water, very broadly linear, with the amount of branching largely reduced, and the expanded branches very regularly arranged.

Form and Branching as Specific Characters.—Some allusion has been made in the foregoing pages to the value of such characters as form and branching of the erect frond for distinctive purposes in describing a species. The question was raised then as to the advisability of making too narrow limits for a species upon such distinctions as form and branching, without regard to the effect of environmental factors. The remarkable variations in these two respects existing within the limits of a single species emphasize this fact, and further call attention to the influence of environmental relations upon form.

Stalk.—The stalk, which characterizes many species, varies in length, width and thickness. The plant, by variations in the length of the stalk, may be sessile, subsessile, or long-stalked. In some forms the stalk is narrow and somewhat thickened; in others it is furnished with a thin, expanded margin. It may vary in thickness from a few cell-layers to a thick, almost fleshy tissue. A midrib, when present, usually runs through the median portion of the stalk. Increase in thickness may take place through a growth of the superficial cells, or through an increase in the number of cells composing the central layer. Frequently the thin margin becomes worn away, and this is accompanied by an increase in thickness of the median portion of the frond, so that the stalk becomes cylindrical.

Midrib.—The midrib presents considerable variation in form and extent. In some species it may be distinguished only as a slight thickening of the median portion of the stalk or frond, while in others it is more highly differentiated, appearing as a ridge of considerable prominence. It

may be either simple and unbranched, as is commonly the case in *N. multilobum*, or it may become considerably divided, as in *N. ruprechtianum*. Frequently it does not extend in the frond beyond the upper portion of the stalk or lower segments. In other cases its ramifications reach out into the branches almost to their tip, and there evanesce, or divide into nerves.

Nerves.—These structures are a characteristic feature of the frond in several species, reaching a high degree of development in some forms. In *N. latissimum* no midrib is present, but the large membranous frond is supported by a network of intersecting nerves and veins of considerable prominence. The other species are not distinguished by such a full development of these structures. Usually the nerves are limited to the outer margins or apices of the frond, where they become flabellate or anastomose freely with each other.

Veins.—More or less conspicuous veins constitute a noteworthy structural element in some species, especially in *N. ruprechtianum* and *N. violaceum*. In these plants, particularly where the frond is at all flabellate, a rich development of the finer venation may be seen, whose ramifications extend in a flabellate fashion throughout the frond, or, in some cases, anastomose with one another, finally becoming free.

In *N. fryeanum*, *N. uncinatum*, and *N. corallinarum* the midrib and conspicuous veins are entirely wanting and the only trace of venation is seen in the microscopic veins which characterize these species. These minute structures are frequently very delicate and invisible to the naked eye. They extend as a rule throughout the frond, branching freely or anastomosing. In *N. fryeanum*, however, the delicate veins, in nearly every case, become somewhat stouter toward the base of the erect frond, where they form a more or less conspicuous fan-shaped area. A single species, *N. spectabile*, is destitute of any sort of veins.

Variable and Fixed Characters.—It has been said in a preceding portion of this paper that stress should be laid

upon the greater value of certain characters as specific distinctions and the less value of others. The more variable characters there mentioned have been sufficiently discussed, and the less variable, more important specific distinctions, based upon the character of the venation and shape and position of the sori, may here be treated.

Venation.—The Californian *Nitophylla* may be separated into groups, distinguished from one another by the character of the venation. In that group which includes the greater number of forms, the species possess a midrib, nerves, and conspicuous veins, developed to a greater or less extent according to the species. Some forms exhibit only the midrib, which passes over at once into the undifferentiated frond as in *N. andersonianum*. Others, again, are provided with a midrib which divides into more or less conspicuous nerves, the latter passing again into the ordinary frond, *e. g.*, *N. harveyanum* and *N. multilobum*. Still other forms, such as *N. violaceum* and *N. ruprechtianum*, show a full development of midrib, nerves, and veins, the last named structures usually conspicuous, and either anastomosing or remaining free and flabellate. A second group consists of forms in which no midrib is present, but the surface of the frond is marked by a network of reticulate nerves and veins, as in *N. latissimum*. The forms of the third group, including *N. fryeanum*, *N. uncinatum* and *N. corallinarum*, are destitute of midrib and nerves, and are provided with scarcely perceptible, usually microscopic veins, which either anastomose with one another, or remain free. A fourth group comprises forms which wholly lack venation of any kind whatsoever, as *e. g.*, *N. spectabile*.

Sori.—The sporangia of *Nitophyllum* are gathered together into sori of varying shape and size. These latter structures may be employed as reliable specific distinctions in discriminating between species. The Californian plants, taken as a whole, show a considerable range of forms as regards the shape of the sorus, and likewise a considerable variety in its position. Within the limits of certain species, *e. g.*, *N. violaceum* and *N. ruprechtianum*, these variations

are remarkable. Notwithstanding this fact, the variations can be so expressed in terms of size, shape and position that clear distinctions can be drawn between species. In some cases the sporangia occur in narrow lines of varying width arranged in a flabellate fashion on the upper divisions of the frond, extending perhaps from its median portion to the margin—their regular position in *N. harveyanum* and a disposition frequently seen in *N. ruprechtianum*—perhaps only found as a fringe just within the margin, as often observed in *N. ruprechtianum*. Other forms have the sori in linear strips or patches along the margin of the frond, an arrangement best seen in *N. violaceum*. In another species, *N. multilobum*, the sori are irregular in shape, rounded or linear-elliptical, with their longer dimensions extending transversely across the frond upon whose upper segments they are borne. The sori occur in other instances as rounded or elliptical patches borne either singly upon distal lobes, as in *N. andersonianum*, or scattered irregularly over the surface, as in *N. spectabile*, sometimes displaying a tendency to arrange themselves into lines, as is well shown by *N. fryeanum*. In yet other plants the sori are minute, rounded structures, densely aggregated between the anastomosing nerves and veins of the frond, *e. g.*, *N. latissimum*.

Proliferation.—The capacity for proliferation possessed by some species is very great. To such an extent does this phenomenon take place, that specimens are often found in which the primary frond has remained comparatively undivided, and has produced from its margin numerous proliferations which exceed in size the primary frond itself. The favorite point for the production of proliferations is along the margin of the frond, especially after this has been weathered by the waves. In such cases the original frond may be reduced to a narrow strip, which will produce proliferations surpassing in size the original portion of the plant. In some species, particularly in *N. ruprechtianum*, the frond becomes reduced to the midrib; this persists for a longer or shorter period, and proliferates very freely, so that by this process the plant practically becomes

perennial. Some species, again, notably *N. violaceum* and *N. ruprechtianum*, bear marginal proliferations in great numbers, upon which appear the sporangia and cystocarps.

Antheridia and Cystocarps.—The antheridia and cystocarps need be mentioned only in a cursory way, inasmuch as they take no prominent part in the identification of the species. The antheridia first make their appearance as whitish patches distributed rather evenly over the surface of the frond. As they mature, the frond becomes wrinkled or rugose to a marked degree. Antheridial plants have now been found in three species, viz., *N. fryeanum*, *N. latissimum* and *N. spectabile*, in considerable abundance. The cystocarps are conspicuous structures, making their appearance irregularly over the surface. In some species the most weathered or reduced plants seem to be favored in the production of cystocarps; and in this case the latter sometimes occur in clusters of proliferations produced from the surface or worn margins of the frond.

Parasites.—In only one instance have the plants of *Nitophyllum* been found to harbor parasites. As a rule, the species are quite free from epiphytes and parasites.

Gonimophyllum buffhami Batters has been found by the writer (1897, p. 81) upon *N. ruprechtianum*. The plants are a pale pink color, contrasting strongly with the deep lake red of the host plant. The parasite seems to prefer the lower portions of the frond as its habitat, occurring on the *Nitophyllum* near its base much more frequently than elsewhere. It grows in patches, extending in some cases an inch in breadth, while the individual fronds reach a height of ten millimeters. So far, *Gonimophyllum* has been found only upon the tetrasporic *N. ruprechtianum*.

Agardh (1898, p. 88) is of the opinion that *Gonimophyllum* is a monstrous form of *N. laceratum*. He suggests further that *N. laceratum* occasionally may be hermaphroditic, contrary to the normal course of development, and that this monstrous form may be the structure concerned in the phenomenon.

Geographical Distribution of the Genus.—The genus *Nitophyllum* is distributed universally throughout the oceans

of the globe. It has been found in the Arctic and Antarctic oceans, along the shores of the continents bordering upon the Atlantic and the Pacific, in the Mediterranean Sea and the Indian ocean, and in the waters of Australia. The coast of the British Islands is particularly rich in species, while some of the finest examples of the genus come from Australia.

About seventy species are known, of which number ten occur on the west coast of North America. Of the ten species thus credited, eight are limited to California or neighboring shores. A ninth species, *N. uncinatum*, is one of the oldest known forms, and is more widely distributed, being found in the Adriatic Sea, on the coast of Australia, in California, on the shores of Europe, in the upper Atlantic and Mediterranean Sea, and in New Zealand waters. The tenth species, *N. harveyanum*, is an inhabitant of New Zealand coasts.

Habitat.—The species of *Nitophyllum* occur on other algæ and on *Phyllospadix;* on bare rocks exposed to the dash of the waves or protected from their violence; or upon rock surfaces which have become thickly coated with Corallines or various Porifera and Bryozoa; or upon the piles of wharves on the surface of the wood. The plants range from the littoral zone between tide marks to the sublittoral zone and have been dredged at a depth of 12–15 fathoms.

IV. Description and Distribution of Species.

Key to the Californian Species of Nitophyllum.

Frond provided with midrib only, or with midrib and nerves, or midrib, nerves, and usually conspicuous veins.

Frond with midrib only, margin of frond serrate or dentate, sori in rounded patches on distal lobes of frond.

N. andersonianum (32).

Frond with midrib and nerves, but without veins.

Sori forming flabellate lines on upper segments of frond.

N. harveyanum (29).

Sori forming transverse patches on upper segments of frond.

N. multilobum (27).

Frond with midrib, nerves and usually conspicuous veins.

Color a pale violet through bright violet red to purple violet; thin and papery in texture; sori forming narrow flabellate lines on upper divisions of frond, or linear patches along margin, or rounded patches borne on the crispate margin or on marginal proliferations, or appearing in both lines and patches. *N. violaceum* (39).

Color a bright red to blackish red; firm and leathery in texture; sori forming wide flabellate lines on upper divisions of frond, or rounded patches on marginal or surface proliferations, or appearing in both lines and patches... *N. ruprechtianum* (34).

Frond without midrib, but provided with network of conspicuous nerves and veins..... *N. latissimum* (16).

Frond without midrib, nerves, or conspicuous veins, but provided with microscopic veins.

Frond ample, erect, membranous below, branching above; sori forming elliptical patches scattered over surface of frond. *N. fryeanum* (22).

Frond erect, branching throughout, apices of ultimate branches recurved or hooked..... .....*N. uncinatum* (26).

Frond minute, creeping, ultimate lobes becoming free; sori forming rounded patches on free lobes; on *Corallina chilensis* (and other algæ ?).....*N. corallinarum* (24).

Frond destitute of veins, membranous; sori forming elliptical patches scattered over surface of frond.....*N. spectabile* (21).

N. latissimum *J. Ag.*

Epicrisis Floridearum, Contin. Spec. Gen. et Ord. Alg., 1876, p. 464.

Hymenena latissima HARVEY, W. H., Proc. Linn. Soc., (Botany) Vol. VI, 1862, p. 170.

N. latissimum AGARDH, J. G., Bidrag Florid. Sys., 1871, p. 49. FARLOW, W. G., Proc. Amer. Acad. Arts and Sci., Vol. X, 1875, p. 365; Report U. S. Fish Comm. for 1875, p. 695, 1876.

N. areolatum EATON, D. C., Farlow, l. c., p. 695.

N. latissimum AGARDH, J. G., Epicrisis Florid., Contin. Spec. Gen. et Ord. Alg., 1876, pp. 464 and 699. FARLOW, W. G., Proc. Amer. Acad. Arts and Sci., Vol. XII, 1877, p. 238. FARLOW, W. G., ANDERSON, C. L., and EATON, D. C., Algæ Amer.-Bor. Exsiccatæ, No. 68, 1878. HERVEY, A. B., Sea Mosses, 1881, p. 175. ANDERSON, C. L., Zoe, Vol. II, 1891, p. 224. HOWE, M. A., Erythea, Vol. I, 1893, p. 68. MCCLATCHIE, A. J., Proc. So. Cal. Acad. Sci., Vol. I, 1897, p. 358. NOTT, C. P., in Phyc. Bor.-Amer., COLLINS, F. S., HOLDEN, I., and SETCHELL, W. A., Fasc. VII; No. 335, 1897. TILDEN, J. E., American Algæ, Century III, No. 212, 1898. AGARDH, J. G., Contin. Spec. Gen. et Ord. Alg., Vol. III, Pt. 3, 1898, p. 83.

N. macroglossum AGARDH, J. G., l. c., p. 84.

Synopsis.—Frond both prostrate and erect. Prostrate frond creeping, slender, linear, much branched; without rhizoids, midrib, nerves, or veins; margin serrate or toothed.

Erect frond subsessile or shortly stalked, flat, membranous; without midrib, but with numerous, conspicuous nerves and veins; palmately divided or segmented, frequently proliferating; with margin usually entire, occasionally minutely serrate. Segments linear or wedge-shaped, usually expanded and lobed at apices. Stalk, when present, usually short, occasionally longer, formed by the wearing away of frond on margins at base and subsequent thickening of nerves and remaining portion of frond. Nerves and veins numerous, conspicuous, extending over almost entire surface of frond, evanescent in outer lobes, branching and anastomosing, frond thus conspicuously areolate.

Proliferations usually numerous, especially upon more reduced portions of frond, membranous, segmented or lobed, nerved and veined.

Sporangia in minute sori scattered over both surfaces of the frond between the nerves and veins, in the conspicuous areoles.

Antheridia forming dull whitish patches over the entire surface, the latter becoming ridged, indented, or rugose.

Cystocarps numerous, large, irregularly disposed over both surfaces, projecting beyond the surface.

Remarks on the Species.—The color of *N. latissimum* does not change to any extent when dried. In the living state the tint is a deep, rich, lake red, becoming a shade darker in the dried condition. Plants frequently occur which attain a length of 35–40 cm., with the frond divided into numerous segments from 3–5 cm. in width. The species is one of the handsomest and most luxuriant in habit of any of the forms inhabiting the coast.

N. latissimum is characterized by its broadly expanded fronds, divided into large, rounded lobes and segments, and by the extensive network of prominent nerves and veins covering the surface. This conspicuous network gives to the surface a markedly areolated appearance, which serves to distinguish the plants from all others of the genus as represented on Californian shores. The frond frequently proliferates along the margin, thus giving rise to numerous ovate or lanceolate proliferations, which possess a well marked venation and often produce sporangia and cystocarps. Proliferations likewise appear freely from reduced portions of the frond.

The creeping, much branched, claw-like, prostrate frond possessed by *N. latissimum* is an excellent adaptation for the purpose of enabling the plant to retain its foothold upon the rough and seamy piles and rock surfaces which are its habitat. Very little variation has been observed in the prostrate frond; and it seems to be a well defined and specially differentiated structure.

Tetrasporic plants are of the most common occurrence. Sporangia are produced very abundantly over the entire surface, between the nerves and veins, thus adding to the distinctive areolate aspect of the plant. The sori are minute and crowded together, giving the impression of large sori completely filling the areoles. Antheridial specimens have been observed in but one locality. They reach a large size, being among the most magnificent examples of the species. In the early stages of development the antheridia form pale, whitish patches. Later, they become more evident, and when well developed, cause the entire frond to appear dull reddish white. The surface of the frond is then decidedly rugose.

A comparison of *N. latissimum* with a specimen of *N. hilliæ* Grev., distributed by Le Jolis in Algues Marines de Cherbourg, No. 215 (in herbarium W. A. Setchell), shows a striking similarity between the two species in texture, venation, and character of lobes or branches. A series of specimens of *N. hilliæ* in the herbarium of Professor W. G. Farlow shows that this plant varies greatly as to prominence of veins. Careful comparison of *N. latissimum* with strongly veined specimens of *N. hilliæ* brings out a strong resemblance between the two. With the material in hand, however, it is not advisable to do more than point out the possibility that the two species may be identical.

It is desirable to call attention at this point to the latest views of Agardh (1876, pp. 464 and 699, 1898, pp. 83–85) concerning *N. latissimum*.

According to Agardh, *N. latissimum* is a form which was first collected by David Lyall at Vancouver Island, B. C., and described by Harvey (1862, p. 170) under the name

Hymenena latissima. A similar form was received somewhat later by Agardh from Golden Gate, San Francisco Bay, collected by Berggren. Further than this, corresponding forms were issued in the Alg. Amer.-Bor. Exsicc. (1878, No. 68), collected by Dr. C. L. Anderson, at Santa Cruz.

Again, according to Agardh, there is still another form, viz., *N. macroglossum*, which is referred to Californian shores. This plant was collected originally by Berggren also at the Golden Gate, and at first was included by Agardh in *N. latissimum*. Specimens collected by the writer (1897, Fasc. VII, No. 335) at the type locality are said by Agardh to be identical with the plant first referred by him to *N. latissimum* and now included under *N. macroglossum*.

The writer has now examined in the field and herbarium a wide range of plants comprehended under these two species of Agardh. Specimens, collected at numerous localities and at various times in the year along the coast from San Pedro northward to Puget Sound entrance, provide a fair amount of material as a basis for estimating Agardh's species. An especially abundant series of forms was collected by the writer between January and May at Golden Gate, San Francisco Bay, the type locality for Agardh's *N. macroglossum*. This collection included numerous specimens of both the species above referred to as established by Agardh.

A discriminating examination of Agardh's descriptions shows, as far as the writer can determine, that *N. latissimum* differs from *N. macroglossum* in being of a paler color, possibly less luxuriant in habit, with by no means such a well developed system of venation. The nerves and veins anastomose less freely, project above the surface of the frond less prominently, or not at all, as is more commonly the case, and soon evanesce into the frond. The areoles are larger and more conspicuous by reason of the broad, flat aspect of the nerves and veins. The sori are fewer in number and are most numerous about the borders of the areoles.

The range of forms examined by the writer with these points in mind exhibits, it is true, the structural differences pointed out by Agardh. But further, it should be observed that the species here in question, as field studies show, is apparently a form occurring between December and August, reaching its finest development in February and March, and at that time exhibiting the characters of *N. macroglossum*. The forms collected from April to August show the characters of *N. latissimum*, with frequent occurrence of transition forms between the two species.

The evidence collected by the writer in field and herbarium points to the conclusion that *N. macroglossum* and *N. latissimum* are seasonal variations of the same species, that is, *N. latissimum*.

Habitat.—On piles of docks and wharves, or on sloping rock surfaces where the wave action is not violent; lower littoral to sublittoral zone.

Distribution.—At various points along the coast from San Pedro to Vancouver.

Localities.—San Pedro! (Mrs. S. P. Monks); Santa Barbara! (Dr. and Mrs. L. M. Dimmick; Mrs. S. P. Cooper); shores of San Luis Obispo County! (Mrs. R. W. Summers); Pacific Grove! (M. A. Howe; Mrs. J. M. Weeks; C. P. Nott); Santa Cruz! (Dr. C. L. Anderson; Mrs. J. M. Weeks); San Francisco! (G. W. Lichtenthaler); San Francisco Bay entrance (Golden Gate) at Fort Point! (M. A. Howe; W. A. Setchell; W. J. V. Osterhout; C. P. Nott); at Lime Point! (C. P. Nott); Fort Ross! (W. A. Setchell); Klatsop, Oregon, (L. F. Henderson, in Herb.; W. G. Farlow, *fide* W. A. Setchell); Port Orchard, Washington! (J. E. Tilden); Esquimault Bay, Vancouver Island, B. C. (Harvey, in Proc. Linn. Soc., (Botany) Vol. VI, 1862, p. 170; Farlow, Proc. Amer. Acad. Arts and Sci., Vol. X, 1875, p. 365); Puget Sound! (N. L. Gardner).

Nitophyllum spectabile *D. C. Eaton.*

In FARLOW, Proc. Amer. Acad. Arts and Sci., Vol. XII, 1877, p. 245.

Nitophyllum spectabile FARLOW, W. G., Report U. S. Fish Comm. for 1875, p. 695, 1876; Proc. Amer. Acad. Arts and Sci., Vol. XII, 1877, p. 238. EATON, D. C., in Farlow, l. c., p. 245. FARLOW, W. G., ANDERSON, C. L., and EATON, D. C., Algæ Amer.-Bor. Exsiccatæ., No. 67, 1878. HERVEY, A. B., Sea Mosses, 1881, p. 174. ANDERSON, C. L., Zoe, Vol. II, 1891, p. 224. AGARDH, J. G., Contin. Spec. Gen. et Ord. Alg., Vol. III, Pt. 3, 1898, p. 43.

Synopsis.—Frond both prostrate and erect. Prostrate frond thin, linear, creeping, destitute of venation; becoming thickened when weathered; branching irregularly; branches rising into erect fronds.

Erect frond sessile or subsessile, flat, membranous; destitute of venation; irregularly oblong, deeply pinnately lobed, occasionally palmately segmented, sometimes proliferating; margin entire, sinuate, or lobed. Segments linear, lanceolate, ovate, or cuneate, frequently deeply lobed at apices.

Sporangia in elliptical sori, disposed at nearly regular intervals over both surfaces of the frond. Antheridia in whitish patches over entire surface of frond, giving to latter an areolate aspect. Cystocarps numerous, conspicuous, irregularly disposed over both surfaces, projecting beyond the surface.

Remarks on the Species.—This plant retains, when dried, the bright, rosy red hue which characterizes it in the living state. The species is said by Professor Eaton, who established it, to reach a length of 50–60 cm. It is one of the largest and finest species of the coast. The general aspect of the frond is much like that of *N. fryeanum*. It differs from that form, however, in not possessing any kind of venation.

Comparison of *N. spectabile* with *N. ruthenicum* (P. & R.) Kjell. aroused a suspicion that the two forms might be identical. A more careful examination showed that, in specimens of *N. ruthenicum* received from Professor Kjellman, the plants were "obsoletely veined below," as is stated in the description of *N. ruthenicum* (1889, p. 25, Pl. I, figs. 11–12), and as is shown in the figure. *N. spectabile*, on the contrary, is totally destitute of venation. All the evidence at hand demonstrates that *N. spectabile* apparently is a distinct species.

The writer is indebted to Professor W. A. Setchell for the following note upon *N. spectabile*, through the courtesy of Professor W. G. Farlow, who kindly permitted an examination of his specimens of *Nitophylla* in connection with

the preparation of this paper. Professor Setchell says, "Specimens in herbarium of Professor Farlow (ex. herb. Acad. Petrop.) labelled '*Aglaophyllum ruthenicum*, Exp. Lutk. ad litora Americanæ borealis-occidentalis, Ross' are young (about an inch high, with no fruit present), and might be the young plants of *N. spectabile*. In the herbarium of Farlow are also several specimens from St. Paul Islands, in Behring Sea (legit White), which might be *N. spectabile*."

The account of this species was advanced to a great extent by examination of a large collection of material from Mrs. J. M. Weeks, of Santa Cruz, Calif., who made an especial effort to secure antheridial, tetrasporic, and cystocarpic plants. The material thus obtained confirms the conclusion that *N. spectabile* is entitled to rank as a distinct species, as established by Professor D. C. Eaton.

Habitat.—On rocks ? or other algæ, sublittoral to elittoral zone. Dredged in 12–15 fathoms, Monterey Bay, Calif.

Distribution.—Along the coast from Santa Monica northward to Santa Cruz.

Localities.—Santa Monica! (Miss S. P. Monks); Pacific Grove, in Monterey Bay! (C. P. Nott); Santa Cruz! (Dr. C. L. Anderson, *fide* Eaton in Farlow, Proc. Amer. Acad. Arts and Sci., Vol. XII, 1877, p. 245; Mrs. J. M. Weeks).

Nitophyllum fryeanum *Farlow.*

Algæ Amer.-Bor. Exsiccatæ, No. 69, 1878.

Nitophyllum fryeanum HARVEY, W. H., Ner. Bor.-Amer., Supp., 1858, p. 128? (See remarks on species). FARLOW, W. G., Proc. Amer. Acad. Arts and Sci., Vol. X, 1875, p. 365; Report U. S. Fish Comm. for 1875, p. 695, 1876. FARLOW, W. G., ANDERSON, C. L., and EATON, D. C., Algæ Amer.-Bor. Exsiccatæ, No. 69, 1878. HERVEY, A. B., Sea Mosses, 1881, p. 176. ANDERSON, C. L., Zoe, Vol. II, 1891, p. 224. HOWE, M. A., Erythea, Vol. I, 1893, p. 68. MCCLATCHIE, A. J., Proc. So. Cal. Acad. Sci., Vol. I, 1897, p. 358. AGARDH, J. G., Contin. Spec. Gen. et Ord. Alg., Vol. III, Pt. 3, 1898, p. 74.

Synopsis.—Frond both prostrate and erect. Prostrate frond much reduced, flat, membranous, lobed, without venation or rhizoids.

Erect frond sessile or subsessile, flat, membranous, with microscopic veins; ---dichotomously or palmately branched or segmented; margin entire,

serrate or toothed. Segments linear, frequently prolonged, occasionally expanded and lobed at apices. Veins not numerous, extending through the frond, branching and anastomosing.

Proliferations minute or wanting, appearing along the margin of the frond.

Sporangia in small elliptical sori scattered over the entire frond, tending to become arranged into lines. Antheridia in whitish areolate patches scattered over surface. Cystocarps conspicuous, irregularly disposed over both surfaces, projecting beyond the surface.

Remarks on the Species.—The color of *N. fryeanum* is very attractive, being a bright rosy red in both the living and dried states. Some of the plants reach a height of 15 cm. The size and more especially the shape is subject to considerable variation. The frond may be short and deeply lobed, or long and branching, with the branches considerably prolonged and linear.

N. fryeanum is noteworthy as being one of the three forms of the coast which possess delicate, microscopic veins. With the exception of this character, *N. fryeanum* and *N. spectabile* have many points in common. The last named species, however, is destitute of any kind of venation.

It is a matter of some doubt whether Harvey's (1858, Supp., p. 128) original description of this plant does not better apply to *N. multilobum*. His mention of a lobed and crenulate margin does not seem to hold good for *N. fryeanum*. Harvey's name, however, was applied to the specimens issued in the Alg. Amer.-Bor. Exsiccatæ (1878, No. 69) and Agardh (1898, p. 74) also retains Harvey's name, with an additional reference to the specimens above mentioned. It seems advisable, therefore, to retain this name for the plant here dealt with, which is identical with that published in the Alg. Amer.-Bor. Exsiccatæ.

There were reasons for supposing that *N. fryeanum* might be referred to *N. ruthenicum* (P. & R.) Kjellman (1889, p. 25, Pl. I, figs. 11–12), with which it agrees to a certain extent in the characters of venation and sori. Carefully selected specimens were forwarded to Professor Kjellman, who replied that the two species were not identical. The plants exchanged with Kjellman for purposes of

comparison, while not wholly inducing the writer to accept Kjellman's conclusion as to the non-identity of the two species, yet do not furnish sufficient reason for declaring them identical. Until a more extended comparison can be made of a wide range of forms, the writer prefers to leave the species as established in the Alg. Amer.-Bor. Exsiccatæ (1878, No. 69).

The writer further takes this opportunity to express his obligations to Mrs. J. M. Weeks for material of *N. fryeanum* collected by her, which permitted a careful study of antheridial, tetrasporic and cystocarpic plants.

Habitat.—No reliable data are at hand concerning the habitat of this species. Some twenty-five specimens seen by the writer were all washed ashore from deep water. The plant presumably occurs upon rocks, and other algæ, in the lower sublittoral, and perhaps elittoral zone.

Distribution.—Known to occur with certainty at but two localities on the Californian coast. At Golden Gate, San Francisco Bay, it apparently has not been collected since the first specimen, if indeed it was identical, was secured by A. D. Frye and forwarded to Harvey.

Localities.—Pebble Beach, Monterey Peninsula! (Miss Bayles); Santa Cruz! (Dr. C. L. Anderson; Mrs. J. M. Weeks); Golden Gate, San Francisco Bay? (A. D. Frye, *fide* Harvey, Ner. Bor.-Amer., Supp., 1858, p. 128).

Nitophyllum corallinarum, sp. nov.

Synopsis.—Frond both prostrate and erect. Prostrate frond creeping, flat, membranous, with microscopic veins and with rhizoids; lobed and branching, with branches becoming erect at intervals, margin entire.

Erect frond subsessile, shortly stalked, flat, membranous, with microscopic veins; ovate-spatulate to elliptical, two to three times longer than broad; subdichotomously lobed or divided, margin entire. Segments minute, ovate, oblong or cuneate. Stalk very short, narrowly linear or cylindrical, passing into a midrib, the latter extending throughout frond, usually branching and free.

Sporangia large, prominent, in sori of varying shape and size, solitary or clustered on body of frond or its segments.

Remarks on the Species.—The form from which the synopsis of this species is drawn up was obtained at San Diego by Mrs. E. Snyder, and sent to the writer by Mr. F. S.

Collins. It is the only specimen which, to the writer's knowledge, has thus far been seen on this coast.

N. corallinarum is a very minute plant in comparison with the other *Nitophylla* of the coast. *Corallina chilensis*, upon which the *Nitophyllum* grows, attains a height between 8 and 12 cm., and in the lower portion a width of 5 cm. or more. The branches of the Coralline are arranged pinnately along the main axis from base to apex of the plant. The general outline is very regularly fan-shaped. The epiphytic *Nitophyllum* extends, by means of its prostrate frond, over the entire surface of the Coralline, to which it adheres firmly by means of the rhizoidal processes that are produced abundantly from the surface in contact with the Coralline. From the prostrate frond rise at regular intervals erect branches which are shortly stalked and expand into ovate-spatulate or elliptical fronds, which may reach a height of 7 mm. and a width of 3 mm. The color is a rosy red to dull carmine. So completely is the Coralline enveloped by the *Nitophyllum*, that the natural color of the former, as well as its jointed structure, is very much obscured. The thin and delicate frond of *N. corallinarum* presents the tessellated surface characteristic of the genus. Throughout both the prostrate and erect fronds may be distinguished minute microscopic veins, which branch more or less freely and remain free.

The sporangia are prominent and large, considering the general minute size of the plant. They occur in clusters rather than in sori of definite shape, and are borne upon the body of the erect frond or on its segments. Tetraspores of unusually large size are formed in the sporangia.

This plant may ultimately be found to be identical with some species of Europe. Such a determination must be left, however, to future observation upon more abundant material.

Habitat.—Epiphytic on *Corallina chilensis.*

Distribution.—Concerning the distribution of *N. corallinarum* little can be said. The Coralline upon which it grows is found along the coast from San Diego northward

to Fort Ross, but no observations are known to the writer on the occurrence of *N. corallinarum* at any other place than the type locality.

Locality.—San Diego! (Mrs. E. Snyder in herb. F. S. Collins).

Nitophyllum uncinatum *J. Ag.*

Spec. Gen. et Ord. Alg., Vol. II, Pt. 2, 1852, p. 654.

Nitophyllum uncinatum McCLATCHIE, A. J., Proc. So. Cal. Acad. Sci., Vol. I, 1897, p. 358; also in Phyk. Bor.-Amer. COLLINS, F. S., Holden, I., and SETCHELL, W. A., Fasc. VII, No. 337, 1897. AGARDH, J. G., Contin. Spec. Gen. et Ord. Alg., Vol. III, Pt. 3, 1898, p. 65.

Synopsis.—Frond both prostrate and erect. Prostrate frond creeping, narrow, linear, thin and membranous; frequently weathered, reduced, and thickened along median portion, with microscopic veins, and with rhizoids; margin entire, or toothed; branching, branches rising into erect fronds at intervals.

Erect frond sessile or subsessile, flat and linear, thin and delicate, sometimes thickened in median portion, with microscopic veins; branching subdichotomously from the base upwards, with margin entire, or occasionally toothed. Branches linear, occasionally expanded, or acuminate, frequently recurved or hooked at apices. Veins microscopic, extending throughout frond, occasionally branching and anastomosing.

"Sporangia in solitary disc-like sori, on the upper branches" or "in rounded sori" on the outer branches. (See Remarks on Species.) Antheridia as yet unobserved. Cystocarps minute, marginal or submarginal, produced at infrequent intervals, projecting slightly beyond surface.[1]

Remarks on the Species.—N. uncinatum has a bright, rosy red tint when alive, usually changing to a dull purplish or brownish red when dried. The fronds may attain a length of 10–15 cm. The plant is one of the more delicate species of the coast, as may be seen in the thin and membranous character of the frond. Throughout the narrow, linear segments extend microscopic veins, which, with the numerous recurved or hooked apices of the branches, may be regarded as the prominent morphological characters.[2]

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[1] Account of cystocarps from specimen in Hauck und Richter, Phykotheca Universalis. Fasc. VII, No. 306, 1889.

[2] Nordhausen (Pringsheim's Jahrbücher f. Wiss. Botanik, Band XXXIV, Heft 2, 1899, p. 263) finds that the hooked apices of the branches of *N. uncinatum* serve as climbing organs.

No fruiting specimens of *N. uncinatum*, so far as can be ascertained, have been reported from Californian shores. The plant seems to be an exclusively southern form on this coast, having been collected only at San Diego and San Pedro, in southern California. From these two localities numerous and abundant collections have been taken, none of which, however, have revealed fruiting specimens. There is a strong probability that the plant propagates itself largely, if not entirely, by vegetative means. It occurs commonly on *Phyllospadix*, in quiet water, conditions of substratum which would favor the active development of the prostrate frond. Its local abundance is shown by its occurrence in such quantities as sometimes to clog the nets of fishermen.

The description of the sori, as given in the synopsis of the species, is taken from Agardh (1852, p. 654, 1876, p. 465, 1898, p. 65). The account of the cystocarp is based upon an examination of the specimen issued in the Phykotheca Universalis (see note under synopsis of species). There seems to be little doubt that the species of this coast is identical with the European plant.

Habitat.—In quiet water, on other algæ, and on *Phyllospadix*.

Distribution.—*N. uncinatum* is a cosmopolitan species, limited in its local distribution, having been found at but two points on the coast.

Localities.—San Diego! (Herb., F. S. Collins); San Pedro! (Mrs. E. A. Lawrence; A. J. McClatchie; Mrs. S. C. Purdy; W. A. Setchell).

Nitophyllum multilobum *J. Ag.*

Epicrisis Floridearum, Contin. Spec. Gen. et Ord. Alg., 1876, p. 698.

Nitophyllum multilobum AGARDH, J. G., Epicrisis Floridearum, Contin. Spec. Gen. et Ord. Alg., 1876, p. 698. FARLOW, W. G., Proc. Amer. Acad. Arts and Sci., Vol. XII, 1877, p. 238; ANDERSON, C. L., Zoe, Vol. II, 1891, p. 224. COLLINS, F. S., HOLDEN, I., and SETCHELL, W. A., Phyc. Bor.-Amer., Fasc. VII, No. 336, 1897. AGARDH, J. G., Contin. Spec. Gen. et Ord. Alg., Vol. III, Pt. 3, 1898, p. 45.

Synopsis.—Frond both prostrate and erect. Prostrate frond creeping, irregularly expanded and membranous, sometimes a rounded expansion, sometimes irregularly branched or lobed; with rhizoids; without midrib, nerves or veins; margin entire, sinuous, or somewhat lobed.

Erect frond slightly stalked, flat and linear, with distinct midrib; simple below, subdichotomously or subpalmately segmented, not proliferating; margin sinuous, toothed or incised. Segments decidedly bullose, usually obtusely lobed, with sinuous or toothed margin. Stalk short, linear, flat, occasionally becoming thickened. Midrib well developed, rather wide, frequently branched, soon evanescent.

Sporangia formed in large, irregularly oblong, frequently lobed or confluent sori, transversely placed upon the segments of the frond. Antheridia as yet unobserved. Cystocarps few, large, conspicuous, scattered over both surfaces.

Remarks on the Species.—In the fresh state *N. multilobum* is a dark red to dull carmine, becoming a burnt carmine to blackish red when dried. The plant is a dwarf one, rarely reaching a height of 9 cm.

The predominant characters of *N. multilobum* are seen in the rather prominent development of the midrib, in the bullose aspect of the frond, and in the peculiar transverse sori. The first named structure is confined to the lower portion of the frond, where it is visible as a definite thickening of the median part, though it does not project prominently above the surface. At its upper extremity it frequently branches, and the resulting portions evanesce very soon into the ordinary tissue of the frond. The bullose frond of *N. multilobum* is an important feature in the appearance of the tetrasporic plant. Usually the surface of the segments which form the upper portion of the plant exhibits this trait. Here the surface is alternately raised and depressed, while the margin becomes crinkled and lobed. The sori, together with the bullose aspect, furnish the most certain means of identifying the species. No other plant of the coast possesses such a characteristic feature as these transverse sori, usually produced in great abundance on the segments of the frond.

The characteristic transverse sori serve to distinguish *N. multilobum* from *N. harveyanum*, with which this plant otherwise has several points in common. In *N. harveyanum*, however, the sori form flabellate lines on the segments of

the frond. In color, the two species are much the same. The midrib of *N. harveyanum* is usually more pronounced than that of *N. multilobum*. *N. harveyanum* is much slenderer than *N. multilobum*, and may attain a height three or four times that of the latter.

Habitat.—On bare rock surfaces or on rocks coated with Corallines, from high water mark to the sublittoral zone.

Distribution.—Limited at the present time to the Californian coast. Has now been reported from Carmel Bay northward to Cape Mendocino. Apparently a northern form.

Localities.—Carmel Bay! (C. P. Nott); Pacific Grove! (C. P. Nott); Santa Cruz! (Mrs. J. M. Weeks); Land's End, San Francisco! (W. A. Setchell; C. P. Nott); Golden Gate, San Francisco Bay (Berggren, *fide* J. Agardh, Epicrisis Floridearum, 1876, p. 698; W. A. Setchell); Lime Point, San Francisco Bay! (C. P. Nott); Dillon's Beach (W. A. Setchell); Fort Ross! (C. P. Nott); Cape Mendocino (C. G. Pringle, in herb., W. G. Farlow, *fide* W. A. Setchell).

Nitophyllum harveyanum *J. Ag.*

Epicrisis Floridearum, Contin. Spec. Gen. et Ord. Alg., 1876, p. 462.

Nitophyllum harveyanum J. AG., Phyk. Bor.-Amer. COLLINS, F. S., HOLDEN, I., and SETCHELL, W. A., Fasc. XIV, No. 693, 1900.

Synopsis.—Frond both prostrate and erect. Prostrate frond creeping, linear, flat; without rhizoids, destitute of midrib, and not proliferating; branching, branches becoming erect at intervals; margin entire, serrate, or somewhat laciniate.

Erect frond stalked, flat, linear; with midrib and flabellate nerves; branching, rarely proliferating, margin entire, or somewhat laciniate. Branches palmate or subpalmate, linear or becoming expanded, occasionally lobed or cleft. Stalk flat, linear, with distinct midrib, becoming thickened and cylindrical through wearing away of margin and renewed growth of median portion. Midrib narrow, conspicuous, branching above, becoming divided into flabellate nerves, the latter conspicuous, branching freely, remaining free and flabellate. Veins minute or wanting.

Sporangia in linear sori extending flabellately from the nerves to the margin of the frond. Antheridia as yet unobserved. Cystocarps large, prominent, irregularly disposed, projecting beyond the surface.

Remarks on the Species.—N. harveyanum varies in color in the living state from deep salmon red to dull carmine, becoming purplish to blackish red when dried. The plant may reach a height of 20 cm., but is as a rule 6–10 cm.

The prostrate frond in *N. harveyanum* does not develop so extensively as in other species. It is destitute of midrib and nerves, and is not specially thickened. *N. harveyanum* is a sparingly branched form as regards its erect frond and the whole plant is rather stiff and unyielding, even when freshly taken from the water. The midrib is conspicuous, tapering slightly towards its upper extremity, and rather suddenly becoming divided into flabellate nerves. The margin in the lower portions of the frond and upon the stalk frequently wears away, the remaining median portion then becoming thickened and cylindrical. In the branches the margin occasionally is serrate or laciniate.

N. harveyanum was first collected on this coast at Land's End, San Francisco, by Professor W. A. Setchell. It grew in company with *N. multilobum*, to which, at this locality, it bears some resemblance, on account of its size and venation. Professor Setchell, however, upon noting the non-bullose character of the frond and the flabellate arrangement of the sori, so different from the transverse sori of *N. multilobum*, concluded that the plant was a distinct species. The writer, when examining the plant in connection with other material secured by him at Fort Ross, was of the opinion that it must be the plant described by Agardh (1876, p. 699) under the name *N. flabelligerum*, although previously the conclusion had been reached that Agardh's *N. flabelligerum* was but a form of *N. ruprechtianum*.

Sufficient comment has been made already upon the distinctions to be drawn between *N. harveyanum* and *N. multilobum*. It is desirable, however, to point out here some of the differences existing between *N. harveyanum* and certain forms of *N. ruprechtianum*. There is enough of similarity between certain variations of the latter species and *N. harveyanum* to give reason for the suspicion that the two are identical. The examination of a good range of

specimens of *N. ruprechtianum* soon brings to light the variation in that species, however, and helps to establish its non-identity with *N. harveyanum*.

Certain forms of *N. ruprechtianum* exhibit a pronounced dark purplish red tinge, both in the fresh and dried states, and are somewhat stiff and brittle. The segments of the frond are narrower, and more or less prolonged. Such forms almost invariably bear flabellately arranged, linear sori, and on the whole, present the distinctive characters of *N. harveyanum*. Between such an extreme variation as this and the typical *N. ruprechtianum*, however, there may be found every gradation in color, form of segments, and position and shape of sori, which are discussed more in detail under *N. ruprechtianum*. The form in question, however, usually retains enough of the distinctive color and venation of *N. ruprechtianum* to enable it to be recognized.

The writer is further indebted to Professor Setchell for a comparison made by him between specimens of *N. harveyanum* in the herbarium of Professor Farlow, and plants from this coast. The specimens in Professor Farlow's herbarium are from New Zealand, and are designated as *N. harveyanum* by Agardh. The resemblance in habit between these and plants collected by the writer at Fort Ross is very striking, both in the tetrasporic and cystocarpic plants. Judging from Agardh's description and from this comparison of specimens, there seems to be good reason for keeping this species under *N. harveyanum*.

Habitat.—*N. harveyanum* is found most frequently upon very much exposed rock surfaces which are bare or coated with Corallines, at extreme low tide-mark in the littoral and sublittoral zones.

Distribution.—Along the coast from Santa Cruz northward to Puget Sound. Apparently a northern form.

Localities.—Santa Cruz! (Dr. C. L. Anderson); San Francisco! (G. W. Lichtenthaler); Land's End, San Francisco! (W. A. Setchell; C. P. Nott); Duxbury Reef! (W. A. Setchell); Fort Ross! (C. P. Nott); Puget Sound! (Thomas Stratton).

Nitophyllum andersonianum *J. Ag.*

Epicrisis Floridearum, Contin. Spec. Gen. et Ord. Alg., 1876, p. 474.[1]

Nitophyllum (Neuroglossum) andersonii FARLOW, W. G., Proc. Amer. Acad. Arts and Sci., Vol. X, 1875, p. 365; Report U. S. Fish Comm. for 1875, p. 696, 1876.

Neuroglossum andersonianum AGARDH, J. G., Epicrisis Floridearum, Contin. Spec. Gen. et Ord. Alg., 1876, p. 474.

Nitophyllum andersonii HERVEY, A. B., Sea Mosses, 1881, p. 177.

Nitophyllum (Neuroglossum) andersonii ANDERSON, C. L., Zoe, Vol. II 1891, p. 224.

Nitophyllum andersonii HOWE, M. A., Erythea, Vol. I, 1893, p. 68. MCCLATCHIE, A. J., Proc. So. Cal. Acad. Sci., Vol. I, 1897, p. 358.

Neuroglossum andersonianum AGARDH, J. G., Contin. Spec. Gen. et Ord. Alg., Vol. III, Pt. 3, 1898, p. 122.

Synopsis.—Frond both prostrate and erect. Prostrate frond creeping, slender, linear, without rhizoids; branching irregularly, occasionally proliferating; without midrib, nerves, or veins. Margin beset at regular intervals with spine-like, sometimes recurved, pinnate teeth. Branches becoming erect at intervals, expanding into erect fronds.

Erect frond shortly stalked, linear, flat, simple below, branching above, with midrib; margin serrate, dentate, or beset with numerous spine-like, pinnate teeth. Branches subpinnately arranged, linear, or alternate at base and expanding at their apices, usually much prolonged. Stalk linear, flat, with definite midrib and thin margin, the margin sometimes disappearing and the median portion becoming thickened, almost cylindrical. Midrib of varying width, becoming prominent, in some cases thickened, almost cylindrical, branching and evanescent in upper portions of frond.

Sporangia in rounded sori, the latter usually large and conspicuous, at the apices of the upper, sometimes expanded, branches. Antheridia and cystocarps so far unobserved.

Remarks on the Species.—The color of *N. andersonianum* varies from bright red to dull carmine when alive, becoming a burnt carmine in the dried specimens. More often the plant has the darker hue mentioned above. The frond may attain a height of 20 cm.

The prostrate frond is commonly slender and much branched, showing much similarity to the corresponding portion of *N. latissimum*. It is destitute of midrib and nerves, and seldom becomes thickened or broadly linear. The erect frond branches freely, while its divisions exhibit considerable variation in width. In some plants they are very slender, linear, and much divided or branched. In

[1] This plant was here for the first time described. It had, in 1875, been mentioned by Farlow (cf. citations) under the name *Nitophyllum (Neuroglossum andersonii)* J. Ag. ms.

others the segments are quite broad and very regularly pinnately arranged. The cause of this variation apparently may be found in the environment. When exposed to violent wave action the fronds become extensively branched. In comparatively quiet waters, on the other hand, the expanded frond reaches its widest development. The predominant characteristic of *N. andersonianum* is the production, along the margin of both the prostrate and erect frond, of numerous pinnately arranged, spine-like, minute projections or teeth. The midrib, in its normal state, is the slightly thickened median portion of the frond, due to an increase in size of the cells of the central layer. Unless the frond is stimulated to further growth by injury, the midrib remains in this state, and, on reaching the upper branches of the frond, soon evanesces. Under the process of weathering, however, the margin becomes worn away. This seems to incite the cells of the median portion to renewed growth, with the result that the stalk and definite midrib become thickened and almost cylindrical. This species does not commonly proliferate.

The tetrasporangia are as yet the only reproductive structures observed. The antheridia and cystocarps have not, so far as can be learned, been seen in the species.

It seems to the writer desirable to allow this species to remain under *Nitophyllum* rather than to assign it to *Neuroglossum*. The habit, sori, and inner structure furnish evidence for thus placing it. Since it was first reported from this coast, doubt seems to have existed in the minds of writers as to whether it was a *Nitophyllum* or a *Neuroglossum*. The synopses already in existence of the genus *Neuroglossum* and of the present species do not at all permit a definite idea to be gained of the internal structure, and widely varying statements exist as to the position of the sori. But the careful study of an extensive range of specimens has served to establish the writer's belief that for the reasons above mentioned the species can be referred to *Nitophyllum* until a comparison can be made with type specimens.

Habitat.—On other algæ and on rocks covered with Porifera and Bryozoa at low tide-mark in the littoral and sublittoral regions.

Distribution.—*N. andersonianum* has now been observed along the coast from San Pedro northward to Carmel Bay. It has never been recorded elsewhere than from the coast of California.

Localities.—San Pedro! (Mrs. E. A. Lawrence); Santa Barbara! (Dr. L. N. Dimmick; Mrs. S. P. Cooper); shores of San Luis Obispo County! (Mrs. R. W. Summers); Carmel Bay! (C. P. Nott); Pacific Grove! (Mrs. J. M. Weeks; M. A. Howe); Santa Cruz! (Dr. C. L. Anderson, Mrs. Boardman).

Nitophyllum ruprechtianum *J. Ag.*

Bidrag till Florideernes Systematik. Lunds Universitets Årsskrift., Tome VIII, 1871, p. 51.

Nitophyllum ruprechtianum FARLOW, W. G., Proc. Amer. Acad. Arts and Sci., Vol. X, 1875, p. 365; Report U. S. Fish Comm. for 1875, p. 696, 1876. AGARDH, J. G., Epicrisis Floridearum, Contin. Spec. Gen. et Ord. Alg., 1876, p. 470.

Nitophyllum flabelligerum AGARDH, J. G., loc. cit., p. 699.

Nitophyllum ruprechtianum HERVEY, A. B., Sea Mosses, 1881, p. 178. ANDERSON, C. L., Zoe, Vol. II, 1891, p. 223. HOWE, M. A., Erythea, Vol. I, 1893, p. 68. MCCLATCHIE, A. J., Proc. So. Cal. Acad. Sci., Vol. I, 1897, p. 358. TILDEN, J. E., American Algæ, Century III, No. 213, 1898. AGARDH, J. G., Contin. Spec. Gen. et Ord. Alg., Vol. III, Pt. 3, 1898, p. 94.

Nitophyllum marginatum AGARDH, J. G., loc. cit., p. 93.

Nitophyllum farlowianum AGARDH, J. G., loc. cit., p. 95.

Synopsis.—Frond both prostrate and erect. Prostrate frond creeping, flat, narrowly membranous, or linear; with rhizoids; frequently proliferating, lobed or branching; often provided with midrib and nerves; margin entire, cuneate, or lobed; frequently forming offshoots or innovations.

Erect frond stalked, with midrib, nerves, and veins; branching, very often proliferating; margin entire, cuneate, undulate, crispulate, laciniate or lobed, often beset with minute proliferations. Branching subdichotomous or subpalmate, with branches linear and often much prolonged, occasionally alternate below, becoming expanded and cuneate above, frequently divided or lobed. Stalk linear, flat, with definite midrib, very often becoming cylindrical through wearing away of margin and thickening of midrib, frequently twisted by wave action, often persistent and freely proliferating. Midrib

conspicuous, branching, sometimes divided into usually conspicuous flabellate nerves and veins, or unfrequently remaining undivided and evanescent; frequently weathered, thickened, persistent, and proliferating freely from sides and end. Nerves flabellate, free or anastomosing, often conspicuous, or inconspicuous and evanescent, sometimes dividing into minute veins. Proliferations produced very freely, on stalk, on margin of frond or on reduced frond, minute and rounded or large, linear, cuneate, frequently lobed or divided, with midrib and flabellate nerves, often bearing sori and cystocarps.

Sporangia in linear sori flabellately disposed about margin of frond, or in linear or irregular submarginal patches, or upon proliferations abundantly produced along margin of frond or upon its surface. Antheridia as yet unobserved. Cystocarps large, infrequent, projecting, irregularly disposed over both surfaces, or gathered together along the margin, or borne upon marginal or surface proliferations.

Remarks on the Species.—Considerable variation in color may be observed in *N. ruprechtianum*. The plants, when young, are often bright red, becoming dull red or carmine with increasing age. The proliferating fronds often exhibit this change in color. When dried, the plant becomes a deep carmine to blackish red. A length of 20–30 cm. is not uncommon, as the plant is vigorous in its growth. Frequently a large number of abundantly branched fronds develop from a single stalk. Good specimens often form masses 30 cm. in diameter and 30 cm. or more in height.

The prostrate frond in *N. ruprechtianum* is developed rather more extensively than in any other species of the coast. It is usually membranous in character, and may either be undifferentiated to any extent or occasionally be provided with midrib and nerves. Owing to the free development and frequent branching of the prostrate frond, there is formed on the substratum an extensive ramification whose outer ends, by the decay or accidental rupture of the older portions of the frond, become separated, and constitute the starting point for a new frond.

The predominant characters of *N. ruprechtianum* are displayed in the robust habit, the abundant proliferations, and the variety in the position of the cystocarps and sori, and the shape of the latter.

Proliferation takes place more abundantly in *N. ruprechtianum* than in any other of the Californian forms. The frond very commonly becomes worn away by the action of

the waves, and by friction upon other algæ and upon rocks. The portions thus reduced persist for a considerable time and give rise to numerous proliferations which exhibit all the characters of the original frond.

It seems strongly probable that the remarkable degree of variation in the shape and position of the sori in *N. ruprechtianum* has been a fruitful source of error to those who have been called upon to identify the forms belonging to this species. It is difficult, perhaps, without extensive examination of plants on the shore at all seasons, to realize what a diversity of form may be found within the specific limits. Especially is this diversity important in considering the sori, which are usually much employed in establishing specific distinctions.

The examination of abundant material of *N. ruprechtianum* has shown that the plants fall into three groups distinguished by the differences in the method of production of the sori, without regard to other features. In one group the sori are produced on the upper, flabellately expanded segments of the frond, and are arranged in rather wide linear sori, or lines, distinct or occasionally confluent, and extending in a more or less connected fashion from the median portion of the segment to the margin. A second group exhibits these linear sori usually confined, however, to the apices of the segments, while along the margin are produced numerous minute proliferations upon which are borne sori having the form of rounded patches. In a third group, the sori are confined to the marginal proliferations or to the similar proliferations appearing upon both the margin and the surface. While, in general, these variations in the position and shape of the sori are seen on different plants, yet it is of great importance in employing them as specific characters to keep in mind the fact that these three different dispositions of the sori are likewise found on one and the same plant.

A similar habit with respect to the production of sori is seen in *N. violaceum*, where, however, the linear sori are much narrower and the marginal ones are often widely

linear and extend in some cases a considerable distance along the margin. When compared with *N. harveyanum*, which also bears linear sori arranged likewise in flabellate fashion, it is seen that, while the resemblance in the production of the sori is strong, *N. harveyanum* possesses a darker purplish tint, is not so robust in habit, and is destitute of such a well developed system of venation as belongs to *N. ruprechtianum*.

The agreement between *N. ruprechtianum* and *N. violaceum* in the production of sori and venation is much more marked, yet the two can be distinguished by the characteristic violet hue and papery texture of *N. violaceum*, in contrast to the dull red color and leathery texture of *N. ruprechtianum*.

The same variety of position as is seen in the case of the sori may also be observed in the cystocarps. These structures may be borne by *N. ruprechtianum*, either upon the surface, when they are scattered at irregular intervals over the entire surface or form a border just within the margin, or they may be borne singly or several together on surface or marginal proliferations.

The existence of such numerous transition forms between the two extremes of surface and marginal production of the sori, a fact clearly established by a careful examination of material from a long extent of coast, points to the conclusion that within the limits of the species known as *N. ruprechtianum* it is possible to include a wide range of forms characterized by the features already pointed out, and that sufficient grounds do not exist for the establishment of several species among which these forms may be distributed.

The evidence afforded by these transition forms should, therefore, be employed in examining the species established by Agardh (1876, p. 699; 1898, pp. 93–96), viz., *N. flabelligerum*, *N. marginatum* and *N. farlowianum*, which, so far as the writer can determine from the descriptions, have been split off from *N. ruprechtianum*. The characterization of these three species is made to

depend upon differences in texture, branching, venation and sori. In the foregoing pages an attempt has been made to show the amount of variation possible in these characters, and to emphasize their relative value for specific purposes. The numerous collections made between widely distant limits along the coast at various points and at different seasons of the year, as well as all the available material at hand in herbaria, have failed to yield forms which could not be referred to *N. ruprechtianum* as defined in the foregoing account of the species.

A word with respect to the general shape and segmentation of the frond may be added in regard to the variation existing in *N. ruprechtianum*. Forms may be found that tend to become elongated, with much prolonged, narrow, and acute segments. Near to these may be placed forms likewise with the segments prolonged, but flabellately expanded, and with rounded apices. The tendency to expansion is seen to best advantage, finally, in plants that become divided into a few broadly wedge-shaped segments, with these in turn somewhat deeply lobed, with the lobes rounded as in the flabellate type.

Habitat.—*N. ruprechtianum* especially occurs on rocks usually covered with Corallines or Bryozoa and Porifera, among which the prostrate frond attains a rich development, in deep rock pools on gently sloping shores, littoral to sublittoral zones.

Distribution.—Along the coast from San Diego, California, northward to Port Orchard, Washington.

Localities.—San Diego! (D. Cleveland); Point Loma! (Miss Minnie Reed); La Holla! (Miss Minnie Reed); San Pedro! (Mrs. S. P. Monks); Santa Barbara! (Dr. and Mrs. L. M. Dimmick; Mrs. S. P. Cooper); shore of San Luis Obispo County! (Mrs. R. W. Summers); Port Harford! (W. A. Setchell); Avila Beach! (Miss Mabel Miles); San Simeon! (E. Palmer); Carmel Bay! (C. P. Nott); Pacific Grove! (M. A. Howe; Mrs. J. M. Weeks; C. P. Nott); Santa Cruz! (Mrs. Boston; C. L. Anderson; Mrs. J. M. Weeks); Duxbury Reef! (W. A. Setchell);

Dillon's Beach! (W. A. Setchell); Fort Ross! (W. A. Setchell; C. P. Nott); Port Orchard, Washington! (J. E. Tilden).

Nitophyllum violaceum *J. Ag.*

Epicrisis Floridearum, Contin. Spec. Gen. et Ord. Alg., 1876, p. 700.

Nitophyllum laceratum HARVEY, W. H., Ner. Bor.-Amer., Vol. II, 1858, p. 104. FARLOW, W. G., Proc. Amer. Acad. Arts and Sci., Vol. X, 1875, p. 365; Report U. S. Fish Comm. for 1875, p. 695, 1876.

Nitophyllum violaceum AGARDH, J. G., Epicrisis Floridearum, Contin. Spec. Gen. et Ord. Alg., 1876, p. 700. FARLOW, W. G., Proc. Amer. Acad. Arts and Sci., Vol. XII, 1877, p. 238. HERVEY, A. B., Sea Mosses, 1881, p. 180. ANDERSON, C. L., Zoe, Vol. II, 1891, p. 224. HOWE, M. A., Erythea, Vol. I, 1893, p. 68. MCCLATCHIE, A. J., Proc. So. Cal. Acad. Sci., Vol. I, 1897, p. 358. NOTT, C. P., in Phyc. Bor.-Amer., COLLINS, F. S., HOLDEN, I., and SETCHELL, W. A., Fasc. VIII, No. 389, 1897. AGARDH, J. G., Contin., Spec. Gen. et Ord. Alg., Vol. III, Pt. 3, 1898, p. 91.

Nitophyllum stenoglossum AGARDH, J. G., loc. cit., p. 92.

Neuroglossum lobuliferum ? AGARDH, J. G., loc. cit., p. 121.

Nitophyllum violaceum formum crispulum SETCHELL, Phyk. Bor.-Amer. COLLINS, F. S., HOLDEN, I., and SETCHELL, W. A., Fasc. XIV, No. 694, 1900.

Synopsis.—Frond both prostrate and erect. Prostrate frond creeping, broadly linear, or membranous, with rhizoids, branching, without midrib or nerves; margin toothed or laciniate.

Erect frond stalked, flat, linear, occasionally with midrib, with flabellate nerves; subdichotomously or subpalmately divided into numerous segments, in some cases finely laciniate, frequently proliferating; margin entire, finely serrate, crispate, or toothed; segments or branches in some cases broadly obcuneate, in other cases narrow, becoming broadly linear, much prolonged and flabellately expanded at apices or remaining linear. Stalk short, soon merging into the frond, usually without midrib, but with flabellate, sometimes anastomosing nerves extending into the branches; or long, somewhat narrow and thickened, almost cylindrical, and again merging into the flabellately nerved branches. Nerves not very conspicuous, usually extending throughout the frond from base nearly to apex, becoming divided into flabellate, frequently anastomosing veins, the latter soon evanescent.

Sporangia in narrow lines flabellately disposed, occasionally confluent, on upper segments of frond, or placed singly or in clusters along the margin, or upon marginal sporophylls, the latter appearing at intervals or in a dense fringe along the margin. Antheridia as yet unobserved. Cystocarps large, projecting, irregularly disposed over both surfaces, or submarginal, or upon marginal proliferations.

Remarks on the Species.—The color of *N. violaceum* varies from a pale violet through bright violet red to purple violet, in both the living and dried states. The plants attain

a height of 20–25 cm. Though not so robust in habit as *N. ruprechtianum*, still some specimens of *N. violaceum* are vigorous in their growth, branching freely and forming large, handsome plants. The plant is papery or parchment-like in texture, rather brittle when dried, and does not adhere well to paper.

N. violaceum, as before remarked, agrees with *N. ruprechtianum* in many morphological details, such as character of branching, general shape and position of sori, and venation; but it may be distinguished from that species by its different color and texture, and the minute differences in size and shape of the sori.

An extremely wide range of variation may be seen in the amount of dissection which the frond of *N. violaceum* undergoes. On the one hand, there is found a form in which the frond becomes divided from the base into a great number of slender, much prolonged branches, which divide again and again, until finally the apices of the ultimate branches are prolonged in a flabellate fashion sufficiently to show the specific characters of venation and color. On the other hand, there occur forms rather broadly membranous at base, that divide into a few broadly obcuneate segments cleft from the outer edge into narrower portions which are prolonged into lobes, again exhibiting the characters of the species. Between these extreme types may be found intermediate forms that in some cases vary toward the finely dissected frond, in others, toward the broadly membranous frond. Stunted and weathered plants also occur in considerable numbers in late winter and spring. The frond in these instances is occasionally thickened and rather fleshy, while the sori are gathered into clusters of marginal proliferations or remnants of the former margin. Proliferation takes place, though not to the same extent as in *N. ruprechtianum*. The proliferations are, as a rule, small, and of varying size. They usually bear sori.

In common with *N. ruprechtianum*, *N. violaceum* challenges attention by reason of the peculiarities of its sori, which exhibit again the same range of variation in regard

to their shape and position that was observed in *N. ruprechtianum*. There are to be found forms with the sori arranged in flabellate, occasionally confluent lines, differing, however, from *N. ruprechtianum* in the relative narrowness of the lines. Numerous transition forms occur which combine the flabellate linear sori with rounded or linear patches upon the margin. Finally, frequent instances may be found of plants bearing the sori as rounded or semicircular patches along the margin, or upon marginal proliferations.

The same conclusion that was reached in regard to the limits of the species in the case of *N. ruprechtianum* may be applied to *N. violaceum*, for here almost exactly similar conditions exist as to specific characters. The variations are in similar directions. Extended study of the forms from numerous localities has shown that they may be referred along the lines there laid down, which leads to the conclusion that *N. violaceum* is a species with limits sufficiently wide to include all the forms common to the coast.

The two species established by Agardh (1898, pp. 92 and 121), viz., *N. stenoglossum* and *Neuroglossum lobuliferum*, seem, therefore, to the writer, as nearly as can be determined from the descriptions, to be forms of *N. violaceum*.

Habitat.—On rocks covered with Corallines, Bryozoa and Porifera, in sheltered situations or in rock pools on gently sloping shores, and on piles of wharves, in the littoral and sublittoral zones.

Distribution.—Along the coast from San Pedro northward to Fort Ross.

Localities.—San Pedro! (Mrs. Lawrence; Mrs. S. C. Purdy); White's Point! (A. J. McClatchie); Santa Barbara! (Dr. L. N. Dimmick; Mrs. S. P. Cooper); San Simeon! (E. Palmer); Carmel Bay! (C. P. Nott); Pacific Grove! (M. A. Howe; C. P. Nott); Santa Cruz! (Dr. C. L. Anderson); Land's End, San Francisco! (C. P. Nott); Fort Point, San Francisco! (M. A. Howe; W. A. Setchell; C. P. Nott); North Beach, San Francisco! (W. A. Setchell; C. P. Nott); Golden Gate, San Francisco Bay (Berggren,

fide J. G. Agardh, Epicrisis Floridearum, 1876, p. 700); San Francisco! (G. W. Lichtenthaler); Lime Point, San Francisco Bay! (C. P. Nott); Duxbury Reef! (W. A. Setchell); Fort Ross! (C. P. Nott).

By way of summarizing some of the features mentioned in the foregoing account, the following table, showing the distribution of species within Californian limits, is incorporated. It will be seen that ten species occur on the coast. These have now been reported from twenty-eight localities, ranging from San Diego northward to Puget Sound, Washington, and points on Vancouver Island, B. C., embracing twenty degrees of latitude and fourteen hundred miles of coast line. It may be pointed out that there is here a coastal distribution equal to that of the Atlantic shores of Europe and the Mediterranean together.

In the light of the evidence collected from a wide range of material in field and herbarium, the ten species recognized may be considered valid until more extended comparison with European specimens proves their identity with previously described species.

Table of Distribution of Species.

(San Diego, Calif., northward to Esquimault Bay, B. C.)

	N. andersonianum.	*N. coralinarum.*	*N. fryeanum.*	*N. harveyanum.*	*N. latissimum.*	*N. multilobum.*	*N. rupprechtianum.*	*N. spectabile.*	*N. uncinatum.*	*N. violaceum.*
San Diego		*					*		*	
Point Loma							*			
La Jolla							*			
San Pedro	*				*				*	*
White's Point										*
Santa Monica								*		
Santa Barbara	*				*		*			*
San Luis Obispo County	*					*	*			*
Port Harford							*			
Avila Beach							*			
San Simeon							*			*
Carmel Bay	*					*	*			*
Pacific Grove	*				*	*	*	*		*
Santa Cruz	*		*	*	*	*	*	*		*
Land's End				*		*				*
Fort Point					*					*
Golden Gate			*?	*		*				*
North Beach										*
San Francisco				*	*					*
Lime Point					*	*				*
Duxbury Reef				*			*			*
Dillon's Beach						*	*			
Fort Ross				*	*	*	*			*
Cape Mendocino						*				
Klatsop, Oregon					*					
Port Orchard, Wash					*		*			
Puget Sound, Wash				*						
Esquimault Bay, B. C.					*					

The writer is deeply indebted to Professor W. A. Setchell, upon whose suggestion this paper was undertaken, not only for collections of material and the use of his private herbarium, but more especially for his discriminating advice and helpful suggestions throughout the entire work, as well as for a comparison of some coast forms with specimens in the herbarium of Professor W. G. Farlow. Further acknowledgment is made to Professor Farlow, who kindly permitted an examination of the *Nitophylla* in his collection, and to Mr. F. S. Collins and Dr. C. L. Anderson, both of whom courteously forwarded to the writer all the *Nitophylla* contained in their herbaria. Acknowledgment should also be made to Mr. Collins for the specimen from which the description of *N. corallinarum* was drawn up. To Mrs. J. M. Weeks the writer is especially indebted for collections which permitted a much more comprehensive account of certain species, and also for the privilege of examining specimens in her herbarium. The herbarium of the University of California was consulted frequently in the preparation of this paper. Special acknowledgment is due to Professors O. P. Jenkins and G. C. Price for their courtesy in extending to the writer during several seasons the privileges of the Hopkin's Seaside Laboratory at Pacific Grove, California, thus facilitating the examination of a large amount of living material, at different times in the year.

UNIVERSITY OF CALIFORNIA,
BERKELEY,
June, 1900.

LITERATURE AND EXSICCATÆ CITED IN THIS PAPER.

1852. AGARDH, J. G. *Spec. Gen. et Ord. Algarum*, Vol. II, Pt. 2.

1871. ———Bidrag till Florideernes Systematik. *Lund's Universitets Årsskrift*, Tome VIII.

1876. ———Epicrisis Floridearum. *Contin. Spec. Gen. et Ord. Algarum.*

1898. ———*Contin. Spec. Gen. et Ord. Algarum*, Vol. III, Pt. 3.

1878. Algæ Americanæ-Borealis Exsiccatæ. FARLOW, W. G., ANDERSON, C. L. and EATON, D. C.

1891. ANDERSON, C. L. List of California Marine Algæ. *Zoe*, Vol. II, No. 3.

1877. EATON, D. C., in FARLOW. *Proc. Amer. Acad. Arts and Sci.*, Vol. XII, p. 245.

1875. FARLOW, W. G. List of the Marine Algæ of the United States, etc. *Proc. Amer. Acad. Arts and Sci.*, Vol. X.

1876. ———Id. *Report U. S. Fish Comm.* for 1875.

1877. ———On Some Algæ New to the United States. *Proc. Amer. Acad. Arts and Sci.*, Vol. XII, No. 20.

1858. HARVEY, W. H. Nereis Boreali-Americana.

1862. ———Notice of a Collection of Algæ made on the Northwest Coast of North America, chiefly at Vancouver's Island, by David Lyall, in the years 1859–61. *Proc. Linn. Soc.*, (Botany), Vol. VI.

1889. HAUCK und RICHTER. *Phykotheca Universalis*, Fasc. VII.

1881. HERVEY, A. B. Sea Mosses.

1893. HOWE, M. A. A Month on the Shores of Monterey Bay. *Erythea*, Vol. I, No. 3.

1889. KJELLMAN, F. R. Om Beringshafvets Algflora. *K. Svenska Vetensk.-Akad. Handl.*, Tome XXIII, No. 8.

1897. MCCLATCHIE, A. J. *Proc. So. Cal. Acad. Sci.*, Vol. I.

1899. NORDHAUSEN, M. Zur Anatomie und Physiologie einiger rankentragender Meeresalgen. *Pringsheim's Jahrbücher f. wiss. Botanik*, Band XXXIV, Heft. 2.

1897*a*. NOTT, C. P. Some Parasitic Florideæ of the Californian Coast. *Erythea*, Vol. V, No. 9.

1897*b*. ———In *Phykotheca Borealis-Americanæ*. COLLINS, F. S., HOLDEN, I. and SETCHELL, W. A. Fasc. VII.

1897*c*. ———l. c. Fasc. VIII.

1897. *Phykotheca Borealis-Americanæ*. COLLINS, F. S., HOLDEN, I. and SETCHELL, W. A. Fasc. VII.

1900. ———l. c. Fasc. XIV.

1898. TILDEN, J. E. American Algæ. Century III.

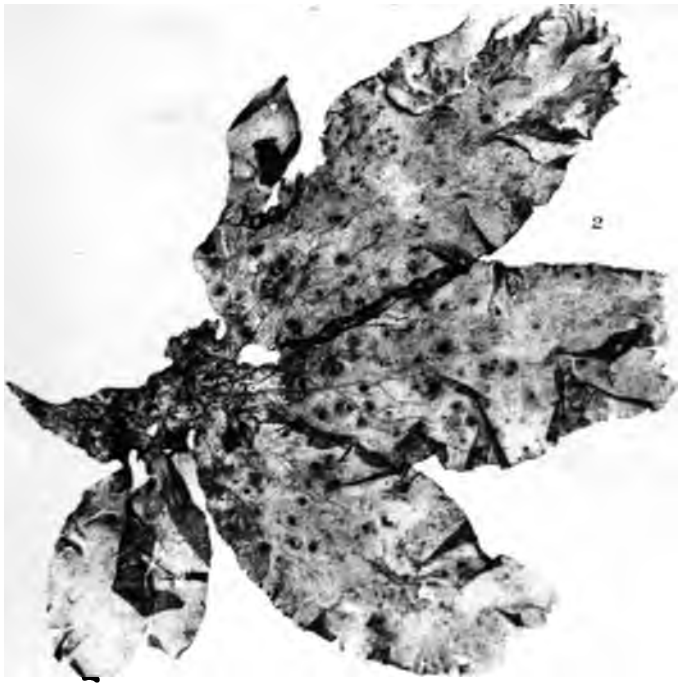
EXPLANATION OF PLATE I.

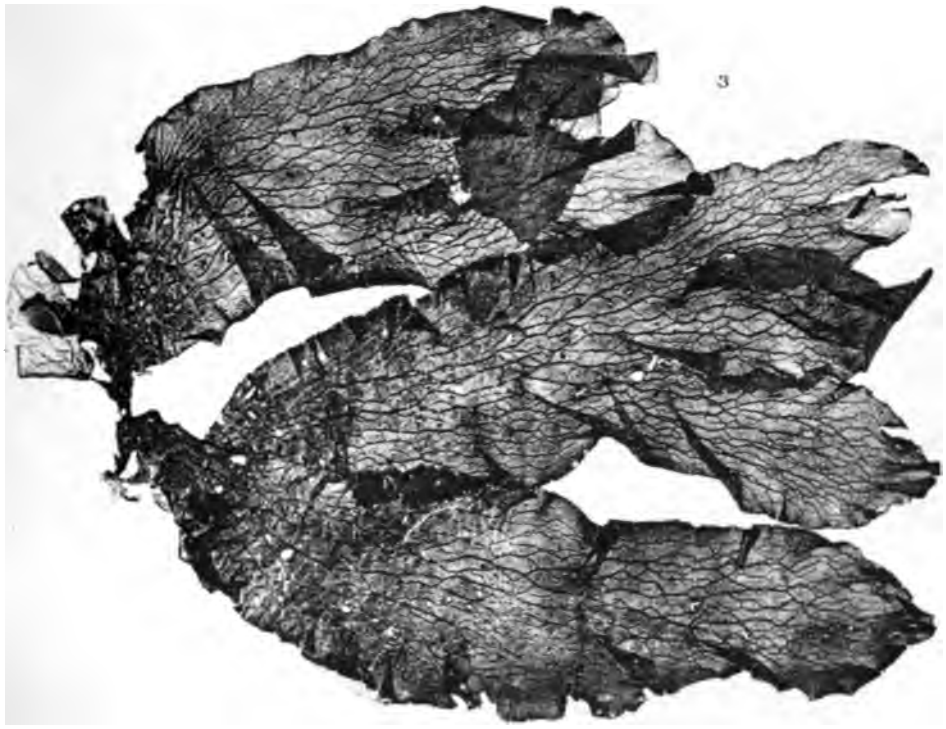
Nitophyllum latissimum.

Fig. 1. Tetrasporic plant of late spring, with broad, flat veins and large areoles. One-third natural size.

Fig. 2. Cystocarpic plant, otherwise as in fig. 1. One-third natural size.

Fig. 3. Antheridial plant, mature, showing in the lower third the characteristic wrinkled surface of the antheridial plant. One-third natural size.





NITOPHYLLUM LATISSIMUM

EXPLANATION OF PLATE II.

Nitophyllum latissimum.

Fig. 4. Typical cystocarpic plant, showing portion of prostrate frond, venation, proliferation, cystocarps, and a portion of the creeping, claw-like prostrate frond. One-third natural size.

Nitophyllum spectabile.

Fig. 5. Typical tetrasporic plant, showing prostrate frond, young and mature erect fronds, and position and shape of sori. One-third natural size.

EXPLANATION OF PLATE III.

Nitophyllum fryeanum.

Fig. 6. Cystocarpic plant, showing disposition of cystocarps and segments of frond. One-third natural size.

Fig. 7. Tetrasporic plant, showing palmate frond with much prolonged segments, arrangement of sori and delicate veins. One-third natural size.

Fig. 8. Tetrasporic plant, showing venation and attenuate segments of frond. One-third natural size.

Fig. 9. Cystocarpic plant, showing pinnately arranged segments of frond. One-third natural size.

Nitophyllum corallinarum.

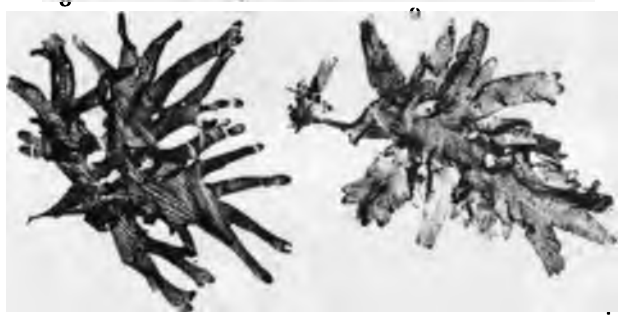
Fig. 10. Tetrasporic plant, showing frond creeping over branches of *Corallina chilensis*, and sori borne on free, erect fronds, *e. g.*, at x and x'. One-half natural size.

Nitophyllum multilobum.

Fig. 11. Tetrasporic plant, showing habit and characteristic transverse sori. One-half natural size.

Nitophyllum uncinatum.

Fig. 12. Sterile plant, showing character of branching and characteristic recurved apices of branches. One-half natural size.



FIGS. 6-9. *NITOPHYLLUM FRYEANUM*

FIG. 11 NITOPHYLL

EXPLANATION OF PLATE IV.

Nitophyllum harveyanum.

Fig. 13. Slender form, tetrasporic, showing habit. One-half natural size.
Fig. 14. Robust form, showing linear sori and flabellately expanded habit. One-half natural size.

Nitophyllum ruprechtianum.

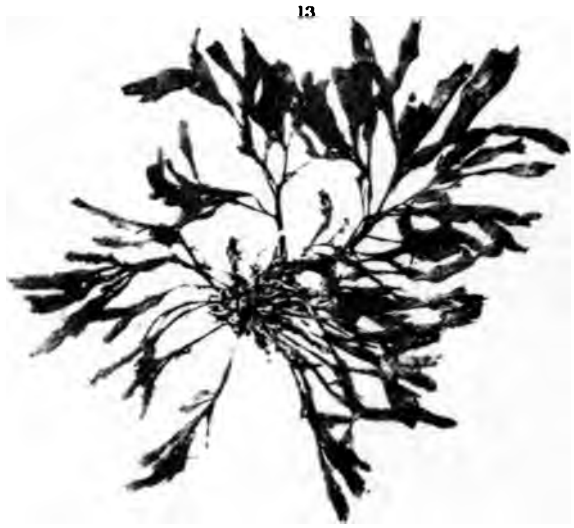
Fig. 15. Prostrate frond of the stout membranous type. One-half natural size.

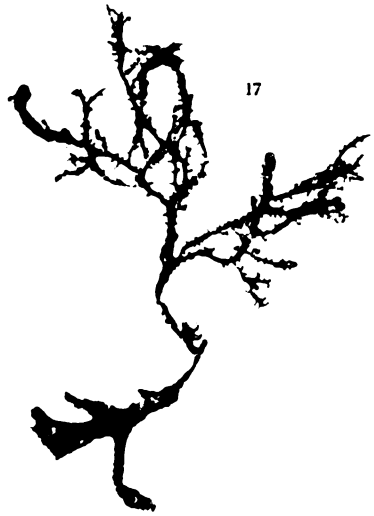
Nitophyllum fryeanum.

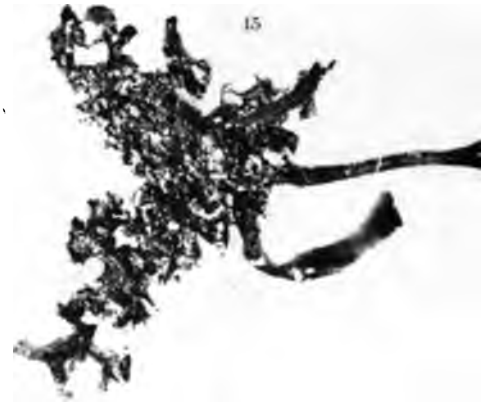
Fig. 16. Prostrate frond of the delicate membranous type, showing rhizoids as minute processes on surface. One-half natural size.

Nitophyllum andersonianum.

Fig. 17. Prostrate frond, of the freely branching type, with portions of the bases of erect fronds. One-half natural size.







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FIGS 13-14. *NITOPHYLLUM HARVEYANUM*.
FIG.15 NITOPHYLLUM RUPRECHTIANUM

FIG.16. NITOPHYLLUM FRYEAN
FIG.17 NITOPHYLLUM ANTIFR-

EXPLANATION OF PLATE V.

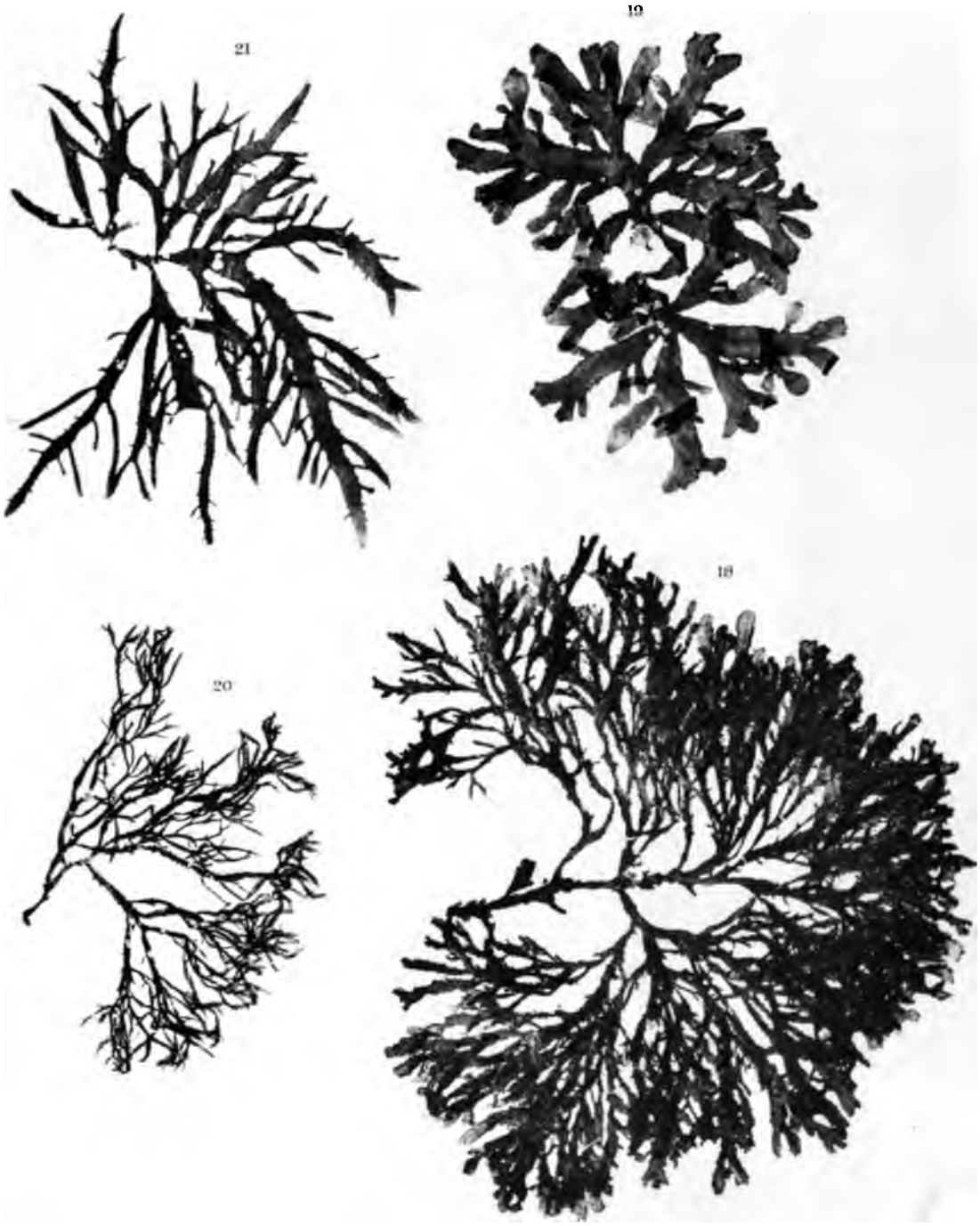
Nitophyllum andersonianum.

Fig. 18. Robust form, sterile, showing usual character of branching, with branches flabellately expanded at apices. One-third natural size.

Fig. 19. Broad membranous form of quiet waters, sterile. One-third natural size.

Fig. 20. Slender form, sterile, showing fine dissection of frond. One-third natural size.

Fig. 21. Transition form, tetrasporic, much branched, showing tendency to become membranous. One-third natural size.



NITOPHYLLUM ANDERSONIANUM

EXPLANATION OF PLATE VI.

Nitophyllum ruprechtianum.

Fig. 22. Expanded, membranous frond, sterile. One-fifth natural size.
Fig. 23. Examples of weathered and proliferating plants. One-fifth natural size.
Fig. 24. Slender, much prolonged type, minutely proliferating along margin, sori on proliferations. One-fifth natural size.
Fig. 25. Typical plant, showing stalk, branching, venation, and proliferations, with sori on proliferations. One-fifth natural size.

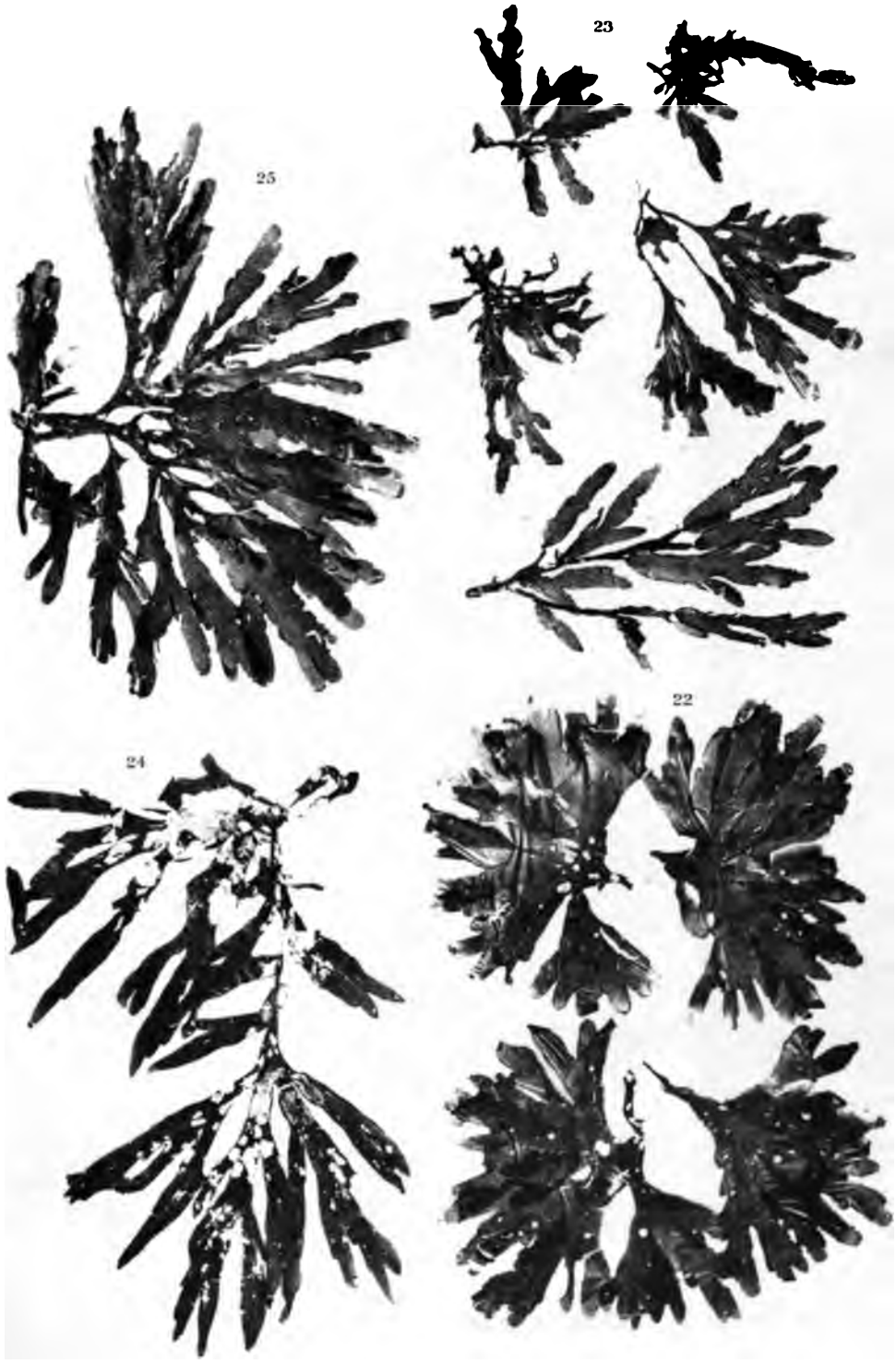


PHOTO LITH PRITTON & W

NITOPHYLLUM RUPRECHTIANUM

EXPLANATION OF PLATE VII.

Nitophyllum ruprechtianum.

Fig. 26. Typical plant, showing stalk, branching, venation, proliferations, and sori on proliferations. One-third natural size.

Fig. 27. Portion of frond, showing flabellately disposed sori. One-third natural size.

Fig. 28. Same, showing sori (linear) confined to apices of branches. One-third natural size.

Fig. 29. Weathered frond, cystocarpic, with prostrate frond. One-third natural size.

Fig. 30. Portion of frond, showing linear sori confined to ends of branches and rounded sori on marginal proliferations. One-third natural size.

26

30

28

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27

EXPLANATION OF PLATE VIII.

Nitophyllum violaceum.

Fig. 31. Intermediate form, showing segments of frond, with crispate margin and marginal sori. One-fifth natural size.

Fig. 32. Typical form, showing slender, linear branches, marginal proliferations, and sori forming linear patches along margin or rounded patches on the proliferations. One-fifth natural size.

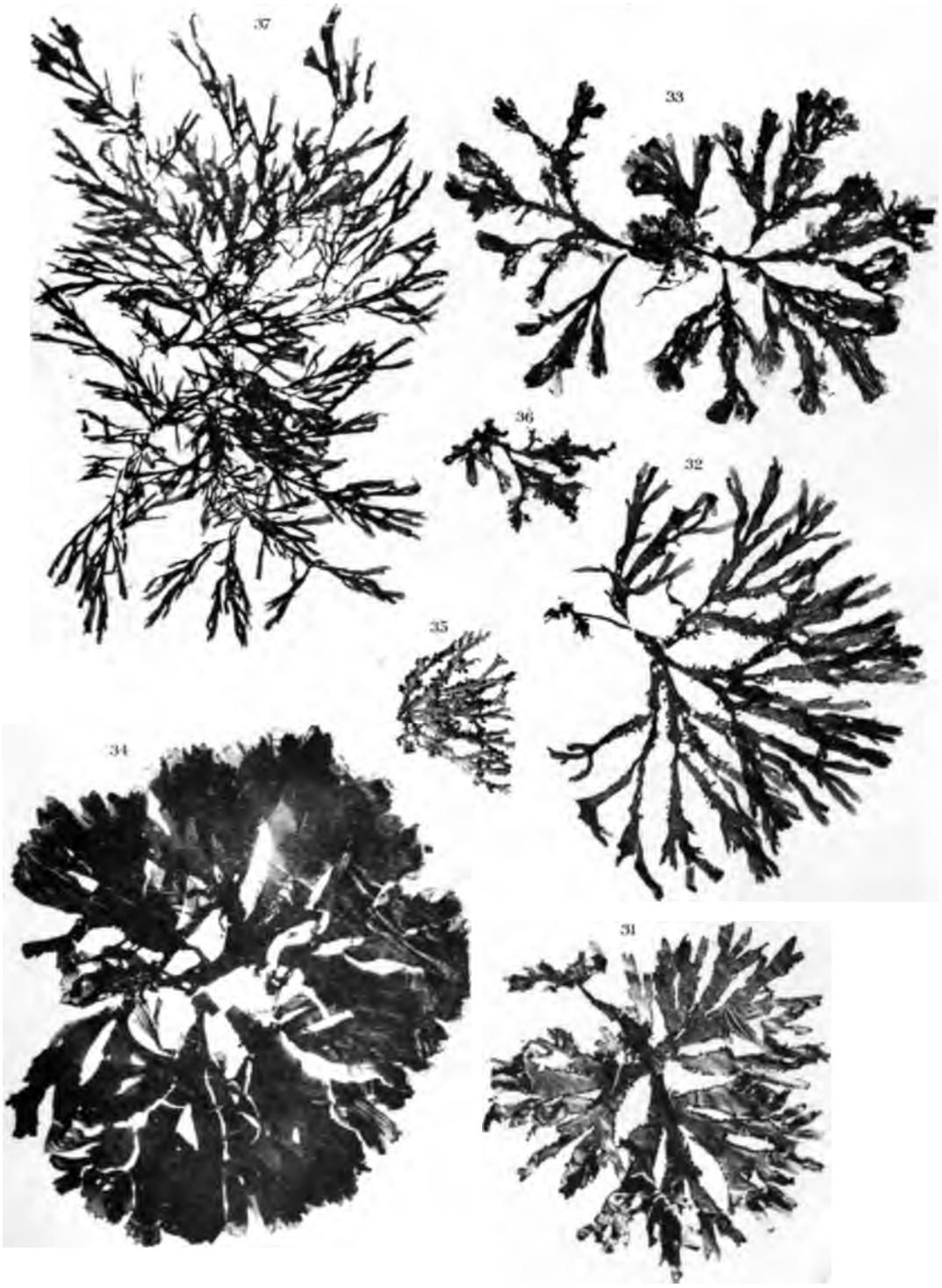
Fig. 33. Intermediate form, with branches flabellately expanded at ends; sori as in preceding figure. One-fifth natural size.

Fig. 34. Robust, membranous form, sterile. One-fifth natural size.

Fig. 35. Reduced and weathered form, the conspicuous sori placed singly or together along the margin, or borne on proliferations. One-fifth natural size.

Fig. 36. Weathered form, as in preceding figure. One-fifth natural size.

Fig. 37. Finely dissected form. One-fifth natural size.



NITOPHYLLUM VIOLACEUM

EXPLANATION OF PLATE IX.

Nitophyllum ruprechtianum.

Fig. 38. Portion of tetrasporic plant, showing rather wide, linear, flabellately disposed sori. Four-fifths natural size.

Fig. 39. Same, showing transition from linear, flabellate sori to rounded sori borne on marginal proliferations. Four-fifths natural size.

Fig. 40. Same, showing sori borne only on marginal proliferations. Four-fifths natural size.

Nitophyllum violaceum.

Fig. 41. Portion of tetrasporic plant, showing narrow, linear, flabellately disposed sori. Four-fifths natural size.

Fig. 42. Same, showing transition from linear, flabellate sori to rounded, inframarginal sori. Four-fifths natural size.

Fig. 43. Same, showing rounded inframarginal sori. Four-fifths natural size.

Nitophyllum latissimum.

Fig. 44. Detail of tetrasporic sori, showing venation, and areoles occupied by minute sori. Four-fifths natural size.

Nitophyllum andersonianum.

Fig. 45. Portion of tetrasporic plant, showing sori as rounded or elliptical patches on distal segments of frond. Four-fifths natural size.

Nitophyllum multilobum.

Fig. 46. Portion of plant, showing characteristic transverse sori. Four-fifths natural size.

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FIGS 32-40 NITOPHYLLUM RUPRECHTIANUM FIG 44 NITOPHYLLUM LATISSIMUM.
FIGS 41-43 NITOPHYLLUM VIOLACEUM FIG 45 NITOPHYLLUM ANDERSONIAN
FIG 46. NITOPHYLLUM MULTILOBUM.

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The Development of the Karyokinetic Spindle in the Pollen-Mother-Cells of Lavatera

BY

EDITH SUMNER BYXBEE

WITH FOUR PLATES

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THE DEVELOPMENT OF THE KARYOKINETIC SPINDLE IN THE POLLEN-MOTHER-CELLS OF LAVATERA.[1]

BY EDITH SUMNER BYXBEE.

PLATES X-XIII.

THE way in which the spindle is formed varies widely in the different families of plants that have been studied. In the generative cells of higher plants, the spindle seems to be always multipolar at first, but the formation of the poles may proceed in several different ways. With a view to shedding further light on this question, the pollen-mother-cells of a species of *Lavatera* were selected for study.[2]

This plant blooms throughout the year, so that the material is plentiful at all times, while the arrangement of the flowers in dense racemes and the number of anthers in a flower make it easy to obtain cells in all stages of division. The pollen-mother-cells are large. They may be examined before preserving, so that much time is saved. The flowers were usually gathered in the morning, as it was found that then the cells were dividing more rapidly. The anthers were examined by crushing them, either with or without the addition of one per cent. glacial acetic acid. All heads in a favorable condition were then immediately dropped into the fixing fluid. It may be mentioned here that all the more striking appearances observed in preserved material were also seen in the fresh. The granular zone, especially, stands out with great distinctness by reason of the strong refractive properties of its granules.

[1] Contributions from the Botanical Laboratories of the University of California, No 13. Presented for the degree of Master of Science. Prepared under the direction of Dr W. J. V. Osterhout.

[2] Owing to the destruction of the plants, it has been impossible to determine the species with exactness. It is probably either *Lavatera unguiculata* Desf. or *L. arborea* L.

The following fixing fluids were tried: Flemming's strong mixture; one per cent. chromic acid; two per cent. iron trichloride; Wilson's corrosive sublimate-acetic; Boveri's picro-acetic; Lindsay's potassium bichromate-platinum chloride-osmic-acetic; one per cent. palladium chloride and one-half per cent. iridium chloride.

Flemming's strong mixture was used undiluted and also with the addition of one, two and three parts of water. The quantities of chromic and acetic acids in the original mixture were also varied. Dilute solutions were found to shrink the cells much more than the strong ones. Varying the amount of chromic acid did not improve the action of the fixing fluid. An increase in the amount of acetic acid, however, gave the best results obtained. Flemming's strong mixture with an excess of acetic acid was therefore almost exclusively used. Fair results were also obtained with palladium chloride and iridium chloride to which a small amount of glacial acetic acid had been added.

After remaining in the fixing fluid for twenty-four hours, the anthers were washed in running water for six hours. They were then placed in a dehydrator[1] for twenty-four hours, with 95 per cent. alcohol below and distilled water above.

Some alcohol was then removed from the material and mixed with an equal volume of 95 per cent. alcohol. The material was transferred to this stronger mixture for two hours. By repeating this process three or four times the material was brought into 95 per cent. alcohol without shrinkage. It was left in 95 per cent. alcohol for twenty-four hours. It was then placed for six hours in each of the following, successively: absolute alcohol; absolute alcohol and bergamot oil (equal parts); bergamot oil; bergamot oil and paraffin, 47° (equal parts); paraffin 47°; paraffin 47° and paraffin 54° (equal parts); paraffin 54°.

Sections 3 to 4 μ in thickness were cut with the Minot wheel microtome. Of the stains tried, Flemming's triple stain (safranin, gentian violet and orange "G") gave the best results.

[1] For a description of the dehydrator see Lawson, 1898, and Williams, 1899.

The pollen-mother-cells are large, and the diameter of the nucleus is equal to fully one-half that of the cell. The chromatin thread is thick and stains blue with gentian violet. The large nucleolus stains red with safranin. It contains a single vacuole. The linin either forms a complete network filling the whole nucleus, or is present as broken threads attached to the chromatin and nucleolus. In neither case does it stain.

The cytoplasm is made up of two constituents, one fibrous, the other granular. The fibrous part forms a network composed of delicate threads crossing each other in every direction. These threads stain deep blue with gentian violet. The other element is composed of small granules, varying somewhat in size.

These granules are scattered throughout the network, both between and upon the fibers, usually in sufficient quantity to give the cytoplasm a cloudy appearance. They have a tendency to collect in small, denser masses at the intersection of the fibers of the network. This tendency is especially noticeable in the earlier stages. A little later the granular matter is distributed more evenly. It stains a brownish yellow with orange "G". A cell in this stage is shown in fig. 1.

The first change that occurs in the cytoplasm is the elongation in a direction parallel to the nuclear wall of the row of meshes immediately surrounding the nucleus (fig. 2). This process goes on until three or four rows of meshes outside the nucleus have become pulled out in this way (fig. 3). These meshes become so long and narrow that, often, on casual inspection, there seem to be threads wound round and round the nucleus. On close examination, however, the meshes can always be seen. Within the nucleus the linin network has meanwhile broken up somewhat; the threads lose their smooth, transparent appearance and begin to stain blue. One or more additional vacuoles appears in the nucleolus, which begins to stain purple rather than red.

Soon after this, the granular substance, which hitherto has been equally distributed throughout the cytoplasm, begins to collect in a denser mass immediately about the nuclear

wall, covering the elongated meshes of the cytoplasmic reticulum (fig. 4). It increases rapidly in quantity about the nucleus without decreasing throughout the rest of the cytoplasm. It is always densest close to the nucleus and from there shades gradually out into the cloudy mass of the cytoplasm.

As this granular substance begins to accumulate, the meshes of the cytoplasmic reticulum, with the exception of a narrow zone surrounding the nucleus, become radially elongated (figs. 4 and 5). This arrangement, however, does not seem to have any particular significance and soon disappears. In a very short time a zone of granular matter has collected about the nucleus, occupying from one-half to one-third of the space outside of it. It becomes so dense that it entirely obscures the reticulum within it, except that a few fibers may sometimes be visible close to the nuclear wall (figs. 5 and 6). By this time, the elongated meshes of the cytoplasm immediately surrounding the nucleus have been transformed into free fibers, which lie between the granular zone and the nuclear wall. Occasionally there is a felt of fibers bounding the outer edge of the granular zone, and, by reason of their deep blue color, standing out conspicuously against the yellowish brown granular matter (figs. 6 and 8). This is by no means constant.

By the time the granular zone is completely formed, the radial arrangement of the reticulum outside of it has entirely disappeared (fig. 6). The granular zone is composed of a dense mass of granules, most of which are larger than those that gave the cytoplasm its cloudy appearance in the earliest stage. In this form it remains unchanged until the end of the anaphase.

While the granular zone has been gathering, the linin in the nucleus has increased somewhat in quantity and in staining power. About this time, the cytoplasm loses its regular structure, as shown in figs. 6, 7, etc. Soon the fibers immediately outside of the nuclear wall come into clearer view, as though the granular matter had withdrawn

from the wall a little, or had been used up at its inner edge (figs. 7 and 8). These fibers, as has already been stated, are probably derived from the elongated meshes of the cytoplasm. They are of much greater diameter than the linin threads, are smooth and stain deep blue. The extent to which these fibers are visible varies greatly in different cells.

At some point the nuclear wall disappears and through the gap thus formed the fibers immediately without the nucleus begin to grow into the cavity (fig. 9). At first, these fibers can be distinguished from the linin threads of the nucleus by their greater diameter and smoother appearance. As the nuclear wall continues to disappear, however, and the linin to thicken up, the fibers from within and without the nucleus mingle in an interwoven mass in which those of different origin cannot be distinguished (fig. 10).

By the time the nuclear wall has entirely disappeared, the nuclear cavity is filled with a mass of interwoven fibers which is usually densest about the circle of chromosomes which marks the situation of the old nuclear wall (fig. 10). In some part of the nuclear space the fibers crowd closer together to form a denser mass and at the same time tend to range themselves so that they lie more or less parallel to each other (figs. 11 and 12). Soon a number of projections appear in this mass as though it were being drawn out at a number of points. Figure 13 shows an earlier and fig. 14 a later stage in this process. This continues until a distinct multipolar spindle is formed (fig. 15).

Soon two principal groups of fibrous cones can be distinguished in the multipolar spindle and in each of these groups one cone becomes the most prominent. Into these two all others are soon absorbed by the continued straightening out and converging of the fibers. Figure 16 shows a spindle in which the cones have almost reached their final position and in which there is only a trace of the third cone. The completion of this process brings the chromosomes into position at the equatorial plate just at the time that the perfect bipolar spindle is formed.

The completed bipolar spindle lies with its poles close against the inner edge of the granular zone, which has assumed an oval shape to accommodate itself to the outline of the spindle. The spindle has sharply pointed ends (fig. 17). It is composed of two sets of fibers. One set runs from pole to pole forming the central spindle. The other runs only from the poles to the chromosomes to which the fibers are attached in bundles. These fibers contract and pull the daughter chromosomes toward the poles. When this process is completed the fibers of the central spindle no longer appear straight but have assumed a wavy appearance. As the daughter chromosomes approach the poles, the mantle fibers appear (fig. 18). About this time the granular zone loses its definite outline. It begins to break up in the plane of the equatorial plate of the spindle and gathers about the poles in two masses in which the daughter chromosomes lie embedded. These two masses remain connected by a shell of granular matter which outlines the old spindle. Figure 19 shows the beginning of this process and fig. 20 a later stage. The cytoplasm seems to contain a greater quantity of granular matter than at any previous time.

Within these two granular masses are formed the daughter nuclei. These have at first a decided indentation on the side toward the spindle (fig. 20). Later they become spherical (fig. 21). The daughter nuclei are thus from the first surrounded by a granular zone which, by the time they are completely formed, has become relatively as wide and as dense as that about the mother nucleus. The central spindle fibers seem to disintegrate and when the nuclei are ready for the second division remain simply as lines of granules connecting the two granular zones (fig. 21).

The second division, as far as could be observed, exactly repeats the process of the first. It was impossible to follow, under the dense granular zone, the elongating of the meshes about the walls of the nuclei, but the concentric lines of fibers were visible in many cases in the narrow space between the zone and the wall. The breaking down of the nuclear wall, the growing in of the fibers, and the formation of the multipolar and bipolar spindles occur as in the first division.

The planes of division of the daughter nuclei do not seem to be at all constant. Sometimes they divide in the same plane, sometimes in planes at right angles to each other, and there are all possible transitions between these two. Figure 22 shows a cell in which the planes are at right angles to each other and fig. 23 one in which they are nearly parallel.

The four daughter nuclei resulting from the second division become surrounded by granular zones just as did the nuclei resulting from the first division. These zones are usually very broad and dense. Connecting them are the mantle fibers across which the cell-plates are formed later on (fig. 24).

The granular zone persists even in the pollen-grain, at least while it is young (fig. 25). It usually occupies at least one-third of the cell space outside of the nucleus. The cytoplasm outside of the zone also contains a great deal of granular matter.

The most important fact in the method of spindle formation above described is that the spindle is formed from free fibers and not from a network. That part of the cytoplasmic reticulum which aids in the formation of the spindle is converted into free fibers at an early stage, long before the nuclear wall breaks down (figs. 5, 6, etc.). The linin network of the nucleus breaks up at an even earlier time. The fibers derived from these two sources become interwoven but never form a true network.

The granular zone, too, is more than usually prominent in *Lavatera*. Its significance will be discussed later.

The higher plants hitherto most exhaustively studied, as *Equisetum* (Osterhout, 1897), *Larix* (Belajeff, 1894), *Cobæa* (Lawson, 1898), and *Passiflora* (Williams, 1899), all show a certain general resemblance to each other and to *Lavatera* in the method of forming the spindle in the reproductive cells. No two of them, however, agree fully in the details. In all of them the first changes in the cytoplasm are either a radial elongation of the meshes of the reticulum or a parallel drawing out of the first two or three rows adjacent to the nucleus.

In *Larix* the radial precedes the parallel elongation, and is of short duration, to judge from Belajeff's figures. In *Equisetum*, *Passiflora*, and *Lavatera* the parallel elongation is the first change observed in the cytoplasm; in *Cobæa* alone it has not yet been observed. In *Larix* and *Lavatera* this condition persists. In the former case, the elongated meshes become part of the central network; in the latter they become transformed into free fibers. In *Passiflora* and *Equisetum* this condition is transitory. In *Equisetum* the parallel elongated meshes are drawn out into an indefinite mass of radially arranged fibers which grow out into the cytoplasm, become parallel to each other, and finally bend together into groups, so that, before its breaking down, the nuclear wall is surrounded by a number of cones. When the nuclear wall disappears, the fibers of these cones grow into the nuclear cavity, come into contact with the linin threads and the chromosomes, and form a multipolar spindle.

In *Passiflora* the radial elongation of the meshes of the reticulum persists for some time. Some of the threads stain more strongly and present an outline suggesting the cones in *Equisetum*, though they are not formed of free fibers. This condition of the cytoplasm is transitory and seems to have nothing to do with the formation of the spindle. This is formed directly from the network resulting from the union of the linin reticulum of the nucleus with that portion of the cytoplasmic reticulum immediately outside of the nucleus. On the breaking down of the nuclear wall these unite to form a continuous network which fills the entire space within the granular zone. The network becomes pulled out at a number of points, and is changed into free fibers which form the multipolar spindle.

In *Larix*, *Cobæa* and *Lavatera*, no cones are present before the nuclear wall disappears. It would seem that the development of these cones is correlated with the slight development of the granular zone. It hardly seems that it would be possible to have such cones in forms like *Cobæa* and *Lavatera*, where the granular zone is very dense. In

Larix, where there is less granular matter, long fibers extend out from the central network—which results, as in *Passiflora*, from the union of cytoplasmic and linin threads—to form by their contraction the cones of the multipolar spindle.

In *Cobæa* and *Lavatera* the spindle formation goes on within the dense granular zone. In *Cobæa* a network is formed as in *Larix* and *Passiflora*. In *Lavatera* the fibers seem to be always distinct and bunch themselves together into a dense mass. The multipolar spindle is formed by a pulling out of the network *(Cobæa)*, or mass of fibers *(Lavatera)*, as in the other cases.

The method of formation of the bipolar from the multipolar spindle does not seem to vary in the various cases.

One of the principal differences, then, in the method of the formation of the spindle in the various plants studied, seems to be the time at which the free fibers are formed from the reticulum of the resting cell. In some cases this occurs very early, as in *Equisetum* and *Lavatera*, which, however, differ widely in other respects. In other cases, as *Larix*, the spindle itself seems to be a network much stretched out.

The granular zone, which is so conspicuous in *Cobæa* and *Lavatera*, has been figured in most of the papers on the division of the generative cells of the higher plants. Osterhout (1897) figures it in the bipolar stage in *Equisetum* and Mottier (1897 *a*) in *Podophyllum* and *Helleborus*. Mottier (1897 *b*) also speaks of its presence at several stages in the divisions in the embryo-sacs of the *Liliaceæ*, but does not seem to regard it as constant in or characteristic of these divisions. Juel (1897) figures it as a prominent ring in *Hemerocallis*, but does not discuss it at any length. In *Passiflora*, also, it forms a well marked zone.

While it is so commonly, perhaps invariably, present in the reproductive cells at the time of their division, it has not been observed in any of the dividing vegetative cells that have been studied. This fact seems to indicate a connection with the two rapid divisions of the reproductive cells for

which it apparently furnishes food material. The granules, too, strongly suggest the yolk granules present in animal eggs. They appear in small quantities in the pollen-mother-cells at an early period, but their exceedingly rapid increase at the time when the first steps in the actual formation of the spindle takes place strongly impresses one with the idea that the cytoplasm is busy producing nutritive material to serve the cell through the period of its activity.

The manner in which the granular zone accumulates suggests the gathering of the deutoplasm in animal eggs (Wilson, 1896, p. 115 et seq.). In *Passiflora* it appears first in patches scattered through the cytoplasm and later gathers into a somewhat loose ring at some distance from the nucleus. This is the way in which the yolk collects in the egg of *Diemyctylus* and other Amphibians, as described by Jordan (Wilson, 1896, p. 116).

In *Cobæa* and *Lavatera* the granular substance appears first close around the nucleus and spreads out from this as in the trout (Henneguy) and cephalopods (Ussow) (See Wilson, 1896, p. 117). The exceedingly dense zone formed by this substance in *Lavatera* exactly resembles that figured by Van Bambeke for a fish *(Scorpæna)* (Wilson, 1896, p. 116, fig. 59, *C*). In some cases, as *Cobæa*, the granular matter is used up as the divisions are completed. In *Lavatera*, however, it constantly increases in quantity up to the formation of the pollen-grains in which it is present, at least in the younger stages. It seems probable that it persists in them and serves them for food during the development of the pollen-tube and the succeeding divisions.

SUMMARY.

1. The cytoplasm of the young pollen-mother-cell is made up of two constituents—a fibrous network and a granular substance.

2. The spindle is formed in the following manner:—

a. The meshes of the network, close to the nuclear wall, pull out in a direction parallel to the wall, forming a felt of fibers about the nucleus.

b. The granular constituent of the cytoplasm collects in a wide, dense zone about the nucleus.

c. The linin increases in quantity.

d. The nuclear wall breaks down and the fibers outside begin to grow into the nuclear cavity.

e. The cytoplasmic and linin fibers form a mass in which the chromosomes lie.

f. The mass of fibers projects out at a number of points, forming the multipolar spindle.

g. Two of the cones become more prominent than the others, which they finally absorb, thereby forming the bipolar spindle.

3. The process in the second division exactly repeats that in the first.

4. The granular substance, forming the dense zone, is comparable with the deutoplasm of animal eggs.

5. Finally, the spindle is formed directly from elements —cytoplasmic and linin reticula—present in the cell from the first, and not from any special spindle-forming substance, or by the aid of centrosomes.

BOTANICAL LABORATORY,
UNIVERSITY OF CALIFORNIA,
October, 1899.

BIBLIOGRAPHY.

1894. BELAJEFF, W. Zur Kenntniss der Karyokinese bei den Pflanzen. *Flora*, Bd. LXXIX, p. 430.

1897. JUEL, H. O. Die Kerntheilung in den Pollenmutterzellen von Hemerocallis fulva, und die bei denselben auftretenden Unregelmässigkeiten. *Pringsh. Jahrb.*, Bd. XXX, p. 205.

1898. LAWSON, A. A. Some Observations on the Development of the Karyokinetic Spindle in the Pollen-Mother-Cells of Cobæa scandens Cav. *Proc. Cal. Acad. Sci.*, 3d Ser., Bot., Vol. I, p. 169.

1897*a*. MOTTIER, D. M. Beiträge zur Kenntniss der Kerntheilung in den Pollenmutterzellen einiger Dikotylen und Monokotylen. *Pringsh. Jahrb.*, Bd. XXX, p. 169.

1897*b*. ———Ueber das Verhalten der Kerne bei der Entwickelung des Embryo-sacks, etc. *Pringsh. Jahrb.*, Bd. XXXI, p. 125.

1897. OSTERHOUT, W. J. V. Ueber Entstehung der karyokinetischen Spindel bei Equisetum. *Pringsh. Jahrb.*, Bd. XXX, p. 155.

1897. STRASBURGER, E. Ueber Cytoplasmastructuren, Kern- und Zelltheilung. *Pringsh. Jahrb.*, Bd. XXX, p. 375.

1896. WILSON, E. B. The Cell. New York. 1896.

1899. WILLIAMS, C. L. The Origin of the Karyokinetic Spindle in Passiflora cœrulea Linn. *Proc. Cal. Acad, Sci.*, 3d Ser., Bot., Vol. I, p. 189.

EXPLANATION OF PLATE X.

All figures were drawn with the Abbe camera lucida: objective, Zeiss oil immersion 1/12, compensating ocular No. 6.

Fig. 1. A young pollen-mother-cell. The chromatin thread is beginning to break up. The cytoplasm is composed of two elements, one a fibrous network, the other a granular substance.

Fig. 2. A little later stage. The first row of meshes adjacent to the nuclear wall has begun to pull out.

Fig. 3. A number of rows of meshes adjacent to the nuclear wall have become elongated. The chromatin thread has broken up.

Fig. 4. The granular substance has begun to collect about the nuclear wall. The outer meshes of the cytoplasm have become radially elongated. The nucleolus shows four vacuoles.

Fig. 5. The granular zone has increased in width. Fibers can be seen between it and the nuclear wall. The linin is thickening up.

Fig. 6. The granular zone is completely formed. The radial arrangement of the meshes outside of it has disappeared.

EXPLANATION OF PLATE XI.

Fig. 7. The space between the granular zone and the nuclear wall has widened and in this space numerous fibers are seen. The cytoplasm has a less regular structure.

Fig. 8. About the same stage as that shown in fig. 7, but there are many more fibers outside the nucleus. There are also strongly staining fibers in the cytoplasm.

Fig. 9. The nuclear wall has disappeared at one point and the fibers outside the nucleus are growing into its cavity. The linin has increased in quantity.

Fig. 10. The nuclear wall has completely disappeared and the space within the granular zone is filled with a mass of interwoven fibers; those near the center of the space are linin.

Fig. 11. The fibers begin to straighten out and arrange themselves in groups, in which they are parallel to each other.

Fig. 12. A more advanced stage in this process.

EXPLANATION OF PLATE XII.

Fig. 13. The fibers begin to converge at a number of points to form cones.
Fig. 14. The cones project out farther from the mass of fibers.
Fig. 15. The fibers have straightened out. The cones are in two groups.
Fig. 16. The cones have nearly fused to form the bipolar spindle.
Fig. 17. A completed bipolar spindle.
Fig. 18. The daughter chromosomes have nearly reached the poles. There are a few mantle fibers. The fibers of the spindle are no longer straight.
Fig. 19. The granular matter is becoming thinner at the equator and collecting more densely about the poles of the spindle. There are traces of radiations which seem to be formed of granular matter extending out from the poles.

EXPLANATION OF PLATE XIII.

Fig. 20. The daughter nuclei are formed. They are at this time crescent shaped as seen in section. There is still a shell of granular matter bounding the remains of the spindle. It is seen in section as two lines.

Fig. 21. The daughter nuclei have become spherical. The granular matter has increased in quantity.

Fig. 22. A spindle of the second division. The other spindle, whose position is indicated by the circular mass of granular matter, lies at right angles to the first.

Fig. 23. Two spindles of the second division that lie nearly parallel to each other. The granular mass between them is not of equal density throughout.

Fig. 24. The four daughter nuclei, each surrounded by a dense granular zone. They are connected by mantle fibers.

Fig. 25. A young pollen-grain. The nucleus is surrounded by a dense granular zone and the cytoplasm outside of this contains much granular matter.

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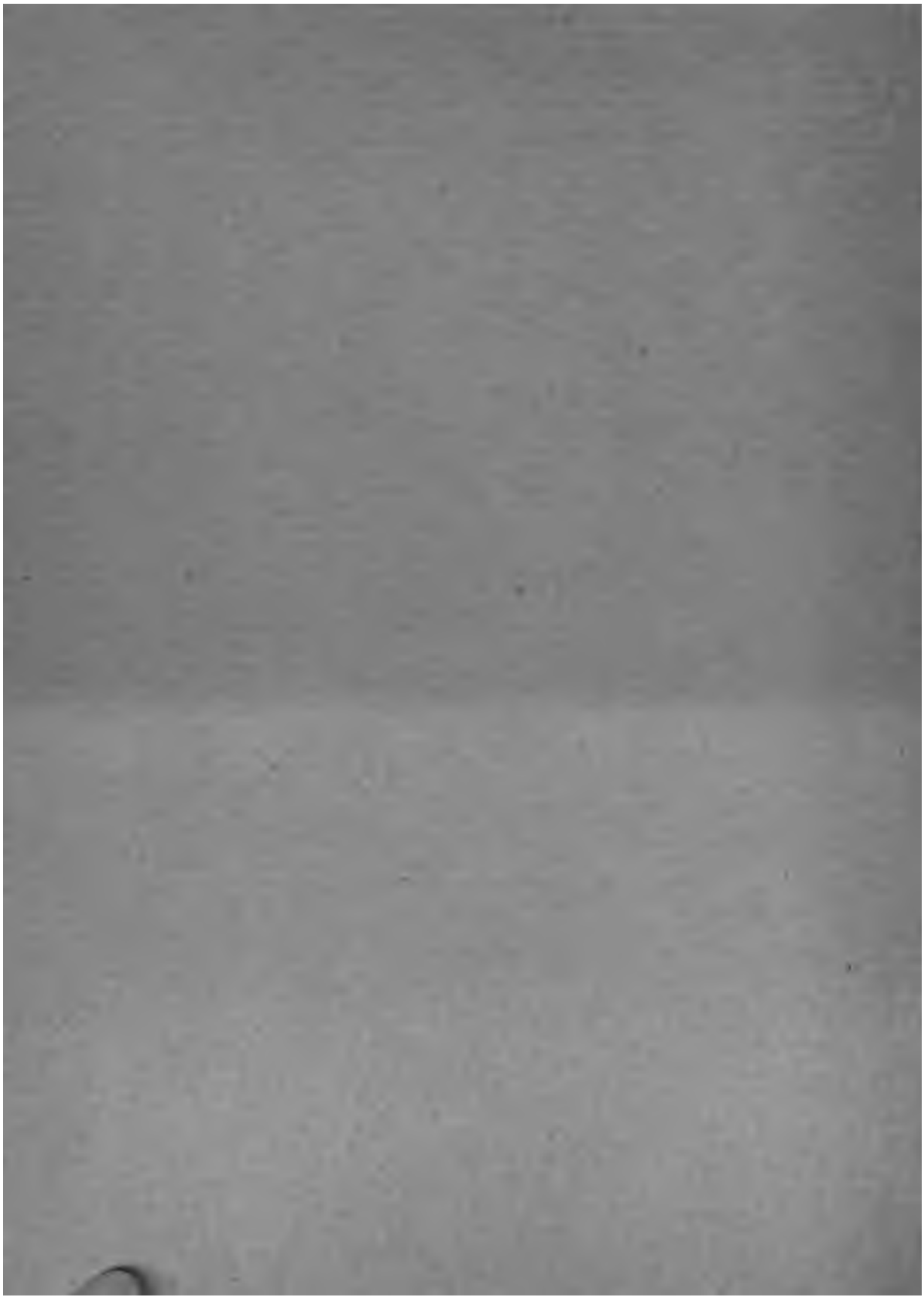

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BOTANY VOL. II, NO. 3

Studies on the Coast Redwood,

Sequoia sempervirens Endl.

BY

GEORGE JAMES PEIRCE

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WITH ONE PLATE

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In the National Geographical Magazine for May, 1899, Gannett asserts that a region naturally forested with redwood will not become reforested with the same tree if the standing timber is felled. He says (l. c., p. 151) "Nowhere is there any young growth. The youngest trees, which are found only in the northern portion of the [redwood] belt, are several hundred years of age. When the timber has been cut, there is no sign of reproduction from seed. In many localities sprouts are starting from stumps in the cut areas, but even this form of reproduction is limited. Indeed, everything seems to indicate that for some reason, probably a progressive drying of the climate, the forest environment is not favorable to the growth of redwood, and that with the clearing away of the present forests, the end of the species as a source of lumber will be at hand." Gannett furnishes a photograph of sprouted redwoods in a cut area.

It is true, as Gannett says, that the major part of the redwood forest is north of San Francisco, especially in Humboldt County, California; but in the Santa Cruz Mountains, south of San Francisco, there are more than merely "scattered groves" of redwood trees. The amount of redwood lumber here cut is evidence that the redwood has attained profitable size and that it still occurs in profitable quantity. Redwood forests must have been abundant in the mountains between the southern arm of the Bay of San Francisco and the ocean, even within comparatively recent years. Much of what was once forested land is now tilled, but by the roadsides and along the fences one sees great blackened stumps which prove the recent presence of redwood forest. In the cañons and on the steeper hillsides, where land cannot under present conditions be profitably cultivated and need not be used for pasture, some of the old redwoods remain. In these same places young redwoods are coming up, some from the stumps, more from the uninjured and still living underground parts of trees which have been felled, and some from seed. So far as this region is concerned, the alarm which Mr. Gannett's remarks arouse seems not to be well founded.

The redwood is a tree of fairly rapid growth. I can not base my opinion on measurements, but I believe that the sprouts from the stumps and underground parts of old redwoods which have been felled grow faster than plants from the seed. It appears, from what I shall presently report, that in the living underground remnants of old trees there are great quantities of reserve food which are available for the nutrition of sprouts. These sprouts or suckers are not wholly dependent upon the food they themselves elaborate. So long as their connection with the parent tree continues unbroken, and the remnants of the old tree retain their vitality, the young trees can use the food stored in the parent. For this or for some other reason, the young trees which begin as suckers and sprouts attain a considerable height and diameter within a few years. Young trees of this sort, in the cañons and on the mountain sides in various places which I have visited, not far from the Stanford University, already have very considerable dimensions. Such trees may be seen above Portola and above Los Gatos. In comparison with the great redwoods of the virgin forest to the north, these young trees are very small. They are still absolutely as well as relatively of no value as timber, but they have been growing only a few years. If they are allowed to continue to grow, if they are reasonably protected against drought by having the watershed above and about them as little disturbed as possible, I can see no reason why valuable redwood timber should not continue to be produced in these cañons at least.

Mr. Gannett's account of the lumbering operations in the northern redwood forest reveals one, and it seems to me an adequate, reason why the redwood forest is succeeded neither by a second growth of seedlings of its own sort nor by a vigorous growth of sprouts from the stumps and underground parts. Because the redwood is free from resin and contains much water, the freshly felled trees do not burn readily. For this reason, the lumbermen commonly clear away the rubbish around the trunk of a felled tree simply by setting fire to the brush. In this way the foliage and

smaller branches are consumed and the main trunk becomes accessible, blackened by the fire, the bark more or less burned through, but the wood uninjured. A fire hot enough to burn up so much green rubbish, though not hot enough to impair the value of the great felled trunks as lumber, is surely hot enough to do great damage to the superficial parts of the stump if not to kill it. Such a fire would probably heat the ground enough and deep enough to injure or kill the underground parts, and it would surely destroy all seeds not deeply buried in the soil. Land cleared in any such way as this usually has to be restocked by plants that wander in, their seeds being blown or brought in by wind, animals, man, etc. It seems to me, therefore, that the habit not of the redwood but of the lumberman is responsible for the failure of the northern redwood forest to renew itself. In view of these facts, is it not unnecessary to imagine any harmful change, if change at all, in the climate of the Pacific Coast since the redwoods have lived here?

Young redwood trees grown from the seed under somewhat artificial conditions often send up suckers from the trunk at or slightly below the level of the ground. A considerable number of redwoods in the Arboretum of Stanford University are doing so. In some instances this may be due to injury to the upper parts of the tree by fungus or animal enemies, but apparently not in all. It seems to me much more likely that the more abundant branching at the bases of these young trees is a reaction to the larger amount of light which falls upon the surface of the soil and upon the lower parts of the trees in the comparatively open Arboretum than in the natural forest. But the Arboretum is not only more brightly lighted than the forest; it is warmer by day and colder by night during some if not all seasons of the year; it receives much less moisture both as rain and as fog than the naturally forest-clad hill and mountain sides; and the soil is not able to retain so much moisture because the ground is naked or nearly so. In the natural forest, unharmed by sheep or man, the ground is covered by a thicker or thinner layer composed of humus, decayed

leaves, leaves only recently fallen, and a great variety of small plants—mosses, lichens, etc.—which form a turf. This covering of the forest floor, as every one knows, is the most important natural means of holding back water, restraining it from too rapid flow, and holding it against evaporation. Some or all of these differences between the natural and the artificial habitat of these trees may act upon them as stimuli to which the production of suckers is the visible reaction.

The Palo Alto, the only large redwood tree still standing on the floor of the Santa Clara Valley, so far toward the Bay of San Francisco, has been subjected to many disturbing influences. Its crown has been seriously injured and its underground parts have been subjected to changed environment. The proximity of the railway embankment and bridge have caused changes in the drainage, both surface and subsoil, and other disturbances less evident must also have occurred. Around the base of this old tree, growing thickly and closely about it, is a brush or thicket of suckers. No young trees have grown up around the parent, forming a little grove such as one sees around the stump of a felled or fallen redwood of advanced age. Only these suckers are formed, close around the trunk, and these are not likely to attain any considerable height or size.

So far as I know, it is only when conditions are unlike those prevailing in the natural forest, or when an old tree has been felled or injured or at least considerably disturbed above or below ground, that it sends up suckers from the trunk or stump, or that young trees come up from the remoter underground parts. These last often make circular groves of greater or less size, known as "redwood temples." Even in the group of giant redwoods at Felton, near Santa Cruz, one sees this arrangement clearly marked. The suckers and sprouts may attain great size in the course of time, as some of these giant redwoods show.

In the production of suckers or sprouts from the trunks and underground parts of *Sequoia sempervirens*, we see the vegetative mode of reproduction engaged in by a species of the Coniferæ; but this recourse to the vegetative mode of

reproduction is probably the result of some external stimulus or stimuli acting upon the plant which, under unchanged conditions, would continue to reproduce itself by seeds if at all. This method of vegetative reproduction is probably sufficient, even under present climatic and other conditions, to secure the continuance of redwood forests in the regions where they now occur, *provided* lumbering operations are so conducted that the production of suckers and sprouts is not made impossible by destructive fires.

II. Peculiarities of Some Vegetatively Produced Young Redwoods.

A. Fasciation.

Fasciation of the young suckers coming up around the trunks of redwoods is not uncommon. The view advocated by Frank (1896), that they are the consequences of an excess of food substances, is strengthened by the time and manner of their appearance. Frank says that fasciations on other plants appear especially when the ordinary branches have been removed or injured in any way. We have seen that the redwoods produce suckers probably only when stimulated to do so by external influences, especially by the removal or at least the injury of the parts above ground. The wound, or other injury, which stimulates the redwood to form suckers, may occur when there is such an abundance of food in immediately usable form, that the production and growth of suckers is so prolific as to insure the fusion of the adjacent parenchymatous parts of the very young branches. In the autumn and early winter, I have had no difficulty in finding fasciated redwood suckers in the Arboretum of Stanford University; they are very noticeable. In the spring and summer months they are by no means so common. I have found no new ones this spring, though there are many young suckers on the redwoods in the Arboretum. Summer, at least the earlier half of the dry season, is the time of food manufacture and storage. In the latter half of the dry season little food can be manufactured because only little water is obtainable. In autumn

the food manufactured and stored becomes available and is used after the first rains have made it possible for growth to be renewed. If, as was the case last winter, there is much mild damp weather, growth will be luxuriant, the stored food will be freely used, the conditions for sucker-formation and for fasciation will coincide. In the spring, especially after a mild winter, during which much growth and comparatively little food manufacture have taken place, the stores of food having been considerably reduced, growth will be less luxuriant and food manufacture will become more necessary and more active. We see, then, some reasons for the formation of the fasciations and for the time of their appearance.

B. Albinism.

The most remarkable and, I am surprised to find, not an especially rare peculiarity of the suckers or sprouts which come up from the stumps or from the old roots of felled or fallen redwood trees, is that they are sometimes perfectly white. My attention was first attracted to this peculiarity when, in the fall of 1898, a student brought some redwood twigs bearing white leaves into the Botanical Laboratory of this University. On inquiry I learned where these white redwoods were growing, and in the fall of 1899 I went to the spot. These are the only white redwoods which I have seen growing, but I have heard of others much larger and one which must be several years old was brought to the laboratory from the "Redwood Retreat," about twelve miles from Gilroy. This last I planted in my garden, but it lived only a short time, whether because it was injured in the transplanting or because it could not bear transplanting late in the spring I do not know. Its behavior before it died I will speak of presently (p. 95).

The white redwoods which I have visited are on the summit not far from the stage road between La Honda and Redwood City, and on the line to the left of the road (as one goes toward La Honda), where the forest gives place to open fields. The tallest redwood tree in view marks the spot where the white ones grow. This tall tree is one of a

number which are several decades old. They, and other smaller young trees of various ages, have come from the stump and roots of a much older tree which must have been very large when it was felled. The old stump has been repeatedly burned under and into, apparently by camp fires, but the heat could not have been enough to do more than local damage, and even this is not great. There is little left of the old stump above the general ground level, but as the hillside falls away abruptly at that point, a good deal of the underground part of the old tree is more or less exposed. Because of the irregularity of the surface of the hillside just there, the old tree sent its roots out more irregularly than is usually the case and the trees which have sprung up from them are not symmetrically placed. There is a thicket, but not a circle or "temple," of redwoods. All of these second growth trees are perfectly normal, as far as I could see. One buttress of the old parent tree, instead of sending up a few more or less scattered sprouts which grow up fairly rapidly and, within a season or two take on the characters of young trees in bark, foliage, and manner of branching, produces branches or bunches of scrubby, thickly set, short and slender sprouts or suckers. These are perfectly white as to leaves. The youngest parts of the stems are of the same ghostly color as the leaves. These white suckers may attain a height of thirty (30) centimeters in the course of one season. They began growing early in April this spring (1900), and they go on growing till hard frost comes. In the same length of time, and with a similar origin, a green sprout or sucker would make two or three or more times this growth in length. The white suckers increase in thickness proportionally to their growth in length, that is, slowly, but the surface of the stem becomes brown and develops cork sooner than the corresponding parts of the green suckers. This precocious cork-formation is not accompanied by other means of protection or by such vigor that the white suckers survive the hard frosts of winter. Even this last winter, milder than usual, was fatal to the white suckers; they were killed down to or just

below the surface of the soil. The green suckers, on the other hand, are enough tougher to survive the winter. In this spot, therefore, white suckers with parts above ground which are two years old are not to be found; but near and just under the ground are well-formed buds which, surviving the winter cold, form the next year's growth of white suckers. In this difference in ability to resist cold, we have one of the physiological differences between the dependent white suckers and the independent green suckers. Whether this is merely a coincidence or a fundamental difference in vigor which forces the white suckers, unable to form chlorophyll, to draw food from the parent if they are to survive, who can tell?

This difference between white and green suckers is not everywhere visible. White redwoods of fair height and age are reported, indefinitely to be sure, from various places in the Santa Cruz Mountains. I have seen white redwoods several years old. These, however, came from places of lighter frosts, if any frost at all touches them during the winter.

Turning now to the anatomy of the white and the green suckers, we see certain peculiarities in the white which demand remark. The leaves evidently present the most marked differences. The leaves of the white suckers are similar in size, form, kind and arrangement to those of the green. There are two kinds of leaves on both green and white suckers—the early or young form, large, long, few, scattered along the stem or branch—and the later or mature form, smaller, shorter, more numerous, regularly placed along the branches, giving to these leafy branches the typical flat and thin dorsi-ventral aspect as compared with the more nearly radial arrangement in the young plant. These two forms, found on the suckers, are also found on seedlings. According to Goebel (1898), the young form is to be regarded as presenting the original leaf-form and leaf-arrangement in the Gymnosperms. The rudimentary characters to be discussed later, which are found in the leaves of the white suckers, would lead one to believe that sooner or later

suckers would be found which retain this primitive character for considerable periods, if not throughout their existence. If Goebel's other supposition is true—that the basal branches, showing the young form of leaves, disappear by correlation as the mature form develops—one would infer the greater physiological perfection and effectiveness of the mature as compared with the young form. But in the white suckers the main function of leaves, that of photosynthetically manufacturing non-nitrogenous food, is entirely suppressed—the mature form of white leaves being as impotent as the young form. Two other important functions of leaves—that of securing and controlling transpiration, and that of securing and controlling the aëration of the plant-body,—would apparently be as perfectly prepared for and accomplished by the one form of leaves as the other, provided the texture and the surface-area of the two kinds of leaves are equal. In these two respects the mature leaves are superior to the young form. There is, therefore, some reason, other than the conception of an "inherited tendency," for the development of the mature form of leaves in the white suckers.

Since the *Sequoias* are geologically such old plants, it is interesting to have the young form of leaves so clearly marked and so constantly recurring. There is a striking resemblance between some of the green suckers in their young condition and the great fronds of *Cycas revoluta*, the leaves of the redwood resembling the leaflets of *Cycas* in form, thickness and arrangement. Can this be a hint as to the origin, perhaps the common origin, of the Coniferæ and the Cycadaceæ?

Comparing the mature form of leaves of green and of white suckers from the same localities, one finds that, despite the superficial likenesses, there are decided structural differences. These are at once evident in cross-sections of the leaves, as shown in figs. 1 and 2. Figure 1 is a diagram of a cross-section of a small green redwood leaf, the single vascular bundle occupying the centre of the leaf, one resin-tube lying under it, the other two resin-tubes

being located at the ends. The upper surface of the leaf is convex, the lower concave, the upper surface and the edges being greatly strengthened by the thickened and heavily cutinized walls of the epidermal cells and by the underlying single layer of sclerenchyma. Beneath this is the very perfectly developed palisade parenchyma, extending from edge to edge of the leaf. The remainder of the mesophyll is composed of simple, unbranched parenchyma, enclosing many intercellular spaces, and bounded on the under side of the leaf by the single layer of fairly thick-walled epidermal cells. In contrast to this, fig. 2, a similar diagrammatic view of the cross-section of a larger white leaf shows a less convex upper, a less concave lower, surface, and the almost or quite complete absence of sclerenchyma cells except at the edges of the leaf (see fig. 9). The most striking difference in the structure of the two leaves, however, consists in the complete absence of palisade parenchyma from the white redwood leaf. The remainder of the mesophyll is composed of somewhat larger parenchyma cells than in the green leaves, and the intercellular spaces are also slightly larger.

Examination of these cross-sections under higher magnification (figs. 7–10) reveals still more plainly the contrast between the white and the green leaves. Figure 7 is a detail from near the middle of the green leaf represented in fig. 1. Figure 7 shows the thick-walled epidermal, the thicker walled sclerenchyma, cells, and the regular palisade-parenchyma cells. These contain many chloroplastids, slightly larger than the starch-grains indicated in the figure. There are many of these starch-grains imbedded in the cytoplasm. Vacuoles are numerous, and evident. The cytoplasm and nucleus are sharply differentiated. Figure 8 represents the corner of fig. 1 cut off by the dotted line. In fig. 8 are shown the very strong sclerenchyma cells immediately underlying the epidermis at the edge of the leaf, and extending almost continuously along the under side to and around the resin-tube. This last is large and bounded by many suitably supported, thin-walled, glandular cells.

At the edges, as well as elsewhere throughout the palisade and mesophyll tissues of the green leaves, starch-grains occur in considerable numbers in all the cells. The amount of starch present undoubtedly varies at different seasons of the year; but since these green leaves were collected at the same time as the white ones, and near them, comparison of the starch content is justified.

Figure 9 represents under higher magnification the part of fig. 2 cut off by the dotted line. As this shows, sclerenchyma cells are found at the edges of the white leaves, but they are not so firm as in the green leaves. The resin-tube is decidedly smaller and bounded by cells evidently less active than the glandular cells of the resin-tubes in the green leaves. There is no starch in any mesophyll cells of the white leaves. The cytoplasm and nucleus are not easily distinguishable from one another in most of the mesophyll cells. The cytoplasm presents a thoroughly disorganized or, as one may more truthfully say, a by no means organized, appearance, neither vacuoles nor plastids being discernible in most cells. The vacuoles occur in few cells only and are unlike those of green mesophyll cells.

The plastids are variable. In sections of white redwood leaves from the summit near where the Redwood City–La Honda stage road crosses the first ridge between here and the sea, I have entirely failed to detect even rudiments of plastids or chromatophores. There are granules and granular aggregations in the cells, as fig. 9 shows, but material carefully fixed in Flemming's weaker mixture of chromosmic-acetic acids and stained by two-tenths per cent. acid fuchsine in distilled water* failed to exhibit any structures which I could positively identify as even rudimentary chromatophores. On the other hand, the material from near Gilroy, treated in exactly the same way, contained chromatophores which ranged in size from those about half as large as the average chloroplastids in the normal green leaves down to indistinguishable rudiments.

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* See Zimmermann-Humphrey, Botanical Microtechnique, pp. 196, 202, etc.

In this connection I may state that the white redwood taken from near Gilroy and planted in my garden had at least one leaf which, after the little tree was transplanted and before it died, became pale green over half its surface on either side of the midrib. I have made no attempt as yet either to transplant, or to disconnect from their parents without transplanting, the white redwoods growing on the summit near the La Honda road. This I shall do presently. Obviously the white redwoods must turn green if they are to survive after being severed from the parent. Some white redwoods can do this more readily than others, the condition of the chromatophores being one of the factors controlling this change. Why there should be these differences in the rudimentary condition of the chromatophores of white redwoods we can understand only after determining the reason for the production of any white leaves at all.

Comparing white redwoods with cedars, which, in cultivation and in nature, not infrequently produce white leaves or green leaves striped or otherwise variegated with white, we find the cell-structure as well as the general conditions for the nutrition of the plants quite unlike. The mesophyll cells in the white parts of green variegated leaves, and of white leaves, contain less protoplasm (Frank, 1896) than do normal green cells, the cytoplasm forming a comparatively thin layer lining the cell-wall, the greater part of the cell-cavity being filled with the more than usually abundant cell-sap. Chromatophores, if visible at all, are colorless, small, and scarcely denser than the cytoplasm, or otherwise distinguishable from it, but they vary in this respect with the degree of whiteness of the leaves. The white redwoods are similar, the cells of the whitest containing no structures recognizable as chromatophores, while those leaves which contain visible chromatophores are not perfectly white. But between the unorganized though abundant protoplasm in the mesophyll cells of the white redwood, and the meagre but very definitely situated protoplasm of the white mesophyll cells of cedar and similar plants, there is a great difference. This we may perhaps account for thus.

Cedars and other plants with white or white variegated leaves are independent, manufacturing in the leaves and other parts that are green the non-nitrogenous foods needed. Any reduction in the number of green leaves, or in the number of chlorophyll-containing-cells in the mesophyll, is a reduction in the capacity of the plant to manufacture food; and if all the leaves turn white the plant will die as soon as the food is consumed which was elaborated and stored while its leaves were green. The turning white, or the failure to become green, of the leaves or any part of the leaves of cedars, etc., is a variation neither useful nor permanent; it is really a morbid condition.

White redwoods, such as I have seen and here describe, are not independent. They absorb from the still living underground parts of the parent tree the non-nitrogenous foods (starch, sugar, etc.) manufactured in its own green leaves and stored in its own underground parts. These stores of food are very great in old redwoods. When for any reason a sucker starts with none of its leaves green, it is exactly as well off, so long as the store of food in its parent lasts, as if its leaves were green and as if it could manufacture food for itself. The activities and possibilities of the white sucker are not abruptly terminated by the exhaustion of its own very limited store of food. It can and does draw on its parent for much food. In the variegated cedar we have some leaves shirking their function as food-manufacturing organs, either because they were defective from the time of their origin at the growing-point, or because they have developed this pathological condition subsequently. The white redwoods, on the contrary, are the vegetatively produced offspring of a wholly independent organism which live as parasites. They take on some of the characters of parasites, as is shown by the absence of palisade cells in the leaves, and by the rudimentary condition of the chromatophores and other protoplasmic contents of the mesophyll cells. They are also less vigorous and grow less rapidly than wholly independent though similarly produced individuals.

Other differences in structure between the white and the green leaves of redwood may be mentioned. As shown by figs. 11, 12 and 13, the walls of the epidermal cells of the white are not as thick as those of the green leaves. Figure 11 shows the epidermal cells around and above the somewhat depressed stoma on a white leaf. Figure 13 is a similar view of a part of the surface of a green leaf. Both figures are of a stoma from the upper surface. Though in the green and in the white the mouth of the stoma is about equal in size, the adjacent epidermal cells are smaller as well as thinner walled in the white redwood. Figure 12 represents the guard-cells of the stoma, only the upper part of which is shown in fig. 11. There are no chromatophores in the guard-cells, but the nuclei are well differentiated.

Though the numbers of stomata on the under side of the green and the white leaves are about equal, there are more stomata on the upper surface of the white leaves than of the green. Figures 3 and 4 represent very diagrammatically the shape and size, but exactly the numbers, of the stomata in equal areas of epidermis from the upper side near the midrib of a green and of a white leaf from redwoods in the Santa Cruz Mountains. Figure 5 indicates the number of stomata in an equal length of epidermis similarly situated but from a green leaf from one of the redwoods in the Arboretum of Stanford University. Figure 6 is another strip from a white redwood leaf from the mountains. Two facts are demonstrated by these figures: first, that the white leaves always have more stomata on the upper side than do the green ones; second, that in the green leaves the number of stomata on the upper side of the leaf is proportioned to the humidity of the region in which the tree grows. The second is a fact well known and understood; the first is new and not easy to understand. In all probability it will be found that the white redwoods occur where transpiration is not so great as in many places in the mountains where green redwoods occur. In a thicket there would evidently be less rapid transpiration than in the open at one side or above the thicket. The white redwood suckers which I have seen are

certainly in a situation in which transpiration can not be very rapid while the suckers are actively growing.

We come now to consider what causes the suckers of certain redwoods to be white while others are green. It is evident that the white redwoods are not white from etiolation, for other suckers similarly situated, and other plants all about, are as green as usual. Nor can lack of iron be the cause, for the same reason. So far as I can see, the only reason for these plants being white is that the leaves form, and attain nearly or quite their full size, at a season when there is insufficient warmth for the formation of chromatophores and chlorophyll pigment, though enough for growth. This is in perfect harmony with Sachs's observation (1864), since extended by Frank (1895), that seedlings growing and buds unfolding at low temperatures produce leaves yellowish or white, either wholly or in patches. I had occasion to notice this phenomenon particularly during the past winter in the leaves of Bur Clover (*Medicago denticulata* Willd). In December and January there were warmth, moisture, and light enough for a lush vegetation composed of the common annuals, but the nights were chilly and the days not warm. There was an unusual amount of variegation in the leaves of the common weeds. That there was light enough for chlorophyll formation is evident from the fact that Bur Clovers growing in the laboratory had no white or variegated leaves. As Sachs proved by experiment, the plant must have a certain minimum amount of warmth in order to form chlorophyll. This minimum, higher than the minimum for growth, of course varies with the species.

The white redwoods growing on the crest near the La Honda road are killed down to the ground each year by the frost. This is evidence of considerable cold. In January of this year (1900), when I visited these white redwoods, they had been frosted down but, buried in the leaf-mould covering the branch of the old stump from which the white redwoods spring, were many buds, healthy, with well formed but perfectly white leaves. On that summit no temperature

records are kept. I therefore cannot tell the temperatures prevailing when these buds were forming in darkness underground. The darkness is of no significance, for all suckers begin in darkness and ordinarily their leaves are then green. Insufficient warmth seems, then, to be the reason why the chromatophores and the chlorophyll pigments do not form in the cells of these growing leaves.

Frank (1895) says that it frequently happens that plants with leaves white because of cold at the time of their formation often retain these white leaves even into the summer, subsequent warmth being insufficient to stimulate to chromatophore and chlorophyll formation. On all such plants, however, the leaves developed later than the white ones, and when the temperature is higher, are green. If this were not the case, the plants would die.

In the white redwood we have a different state of affairs. Although the first leaves borne on a shoot of one year's growth may all have been formed in the cold, late in the previous year, and therefore may not be able to turn green, the leaves later formed, and the internodes forming or at least elongating later, when there is sufficient warmth, one would expect to find green. On the contrary, once started as white redwoods, the suckers continue white as to leaves and young cortex for an indefinite time.

The differences in the color, and in the condition of the chromatophores between the white redwood leaves from near the La Honda road, and those from near Gilroy, may be accounted for thus. It is probable that near Gilroy, at least in the spot where the white redwoods grow, the temperature is not so low as on the exposed summit crossed by the La Honda road. While the temperature is low enough to prevent chlorophyll formation, it is not low enough completely to suppress the formation of chromatophores, and by no means low enough to interfere with growth. Slight variations in low temperatures at the times when the buds, from which suckers spring, are forming, might permit the formation of green buds, of yellowish buds with rudimentary chromatophores, and of white buds with no chromatophores

at all. Certain it is that all the white redwoods I have seen or heard of grow where the temperature is low in autumn and winter.

III. The Significance of the White Redwoods in Connection with our Conceptions of Parasitism and of Heredity.

White redwoods are wholly dependent, absolutely parasitic, plants which are in their first generation. They are not the offspring of other white redwoods, they are not the descendants of a long line of more and more dependent, more and more degenerate, organisms. Their parasitic characters have been acquired, or developed, during the brief course of their own existence, but they possess some parasitic characters not yet acquired by plants which have been semiparasitic for no one knows how long. *Phoradendron*, *Viscum*, and the other "green parasites" have long lived at the expense of the other plants upon which they grow; but though attached to their hosts, these parasites manufacture in their own green leaves their own non-nitrogenous foods.

As I have shown elsewhere (1893), the "green parasites" which have been studied differ from completely parasitic flowering plants (e. g. *Cuscuta*, *Brugmansia*, *Rafflesia*) in the completeness of the connection between the tissues of host and parasite effected by the haustoria. In the complete parasites, xylem and phloëm of the parasite are directly connected with the xylem and phloëm of the host by means of xylem and phloëm tissues which are continuous throughout the haustoria. In the "green parasites," on the other hand, only the xylems of host and parasite are directly connected. This anatomical difference may be considered the reason for the difference in the degree of parasitism in these two sets of plants, or we may conceive that, so long as the parasite remains green, and therefore able to manufacture its own food, a complete connection with both sets of conducting tissues in its host is unnecessary and unformed. There is at present no means to decide which

is true. In the first place, we do not know the actual functions, and cannot determine the entire significance, of the phloëm tissues in higher plants. In the second place, so long as Trelease (1894), or any one else, can say without proof of error, that *Leitneria floridana*, a tree, contains no sieve-plates and inferably no sieve-tubes, the elements of phloëm commonly regarded as most essential, it is impossible to conclude that sieve-tubes are indispensable or that a phloëm as well as xylem connection between host and parasite is essential for complete parasitism. Until the chemical physiology, and not the anatomy only, of the relation existing between parasite and host in *Viscum*, *Phoradendron*, *Cuscuta* is worked out, it cannot be known how significant and important are the tissue connections effected by the haustoria.

As to the significance of these tissue connections, the conditions presented by the white redwood may furnish some idea. The dependent white redwoods are branches of independent parents and are therefore connected from their beginnings, xylem with xylem, phloëm with phloëm, and parenchyma with parenchyma, with their parents. By means of these connections the adequate supply of foods, as well as of food-materials and water, by the parent to its offspring, from old redwood host to parasitic sucker, is assured from the first. On the other hand, a parasite attacking the tree from the outside must establish these connections. It may establish them only imperfectly, as in *Viscum*, or completely, as in *Cuscuta*. The white redwood, with its perfect connection with the parent, offers the counterpart of the condition which accompanies complete parasitism, and though the leaves persist as such, they are structurally no longer perfect leaves, and physiologically only partly so. Because of its perfect connection with the host, the white redwood is able immediately to develop some features of the characteristic structure of parasites.

This is especially interesting because the white redwoods are the vegetatively produced offspring of independent plants, themselves the descendants of generations of independent plants. The suckers of redwoods inherit the

tendency, which one would expect to find firmly fixed, to develop into independent plants, green and manufacturing their own foods. But these young suckers, originating when the weather is too cool for chlorophyll and chromatophore formation, though warm enough for a certain amount of growth, beginning with white leaves and growing well enough during the season without manufacturing food for themselves, form no chlorophyll even in the leaves and internodes later developed. The effect of low temperatures when the first leaves were forming was either upon the protoplasm itself, preventing its forming chromatophores and chlorophyll, or upon the chemical processes by which these organs and substances are produced. One naturally assumes the former—that the powers of the protoplasm are lessened by low temperatures. In these first leaves, the protoplasm is prevented by the cold from following its inherited tendency to produce chromatophores and chlorophyll. In the leaves later formed, the inherited tendency to form chromatophores and chlorophyll is not interfered with by cold, but it does not cause these leaves to become green. They do not need to be green; the plant obtains food enough without turning green and manufacturing its own food. The inherited tendency is not aroused by hunger into action. The stimulus needed to set it in operation not being given, the inherited tendency remains dormant as long as the white suckers remain connected with the parent. The parasitic habit forced upon the young white sucker by its inability to manufacture its own food, and the parasitic characters assumed by the young white sucker, are continued as the plant grows. Continued healthy existence in spite of inability to manufacture food induces in new leaves and cortex those characters found in the earlier and older ones. Environment, the influence of certain stimuli, induce a reaction ordinarily characteristic of species of plants which have been parasitic for generations.

The white redwood serves as an index of the relative powers of heredity and of environment, or, more definitely, of heredity and of the influence of, and the power of

reaction to, certain stimuli. There being no need to manufacture food, the food-manufacturing apparatus is not formed, a parasitic habit being successful so far as the individual is concerned, the inherited habit is not entered upon.

That the need to manufacture food would have an effect upon the development of the white suckers is indicated by the behavior of the white sucker which grew near Gilroy and which I planted in my garden where it could obtain little if any organic matter as food. It died soon after transplanting, but *not until one leaf had become pale green.* The effect of cutting white suckers away from the parent stock and from their supply of manufactured food I shall test presently by experiment on the redwoods near the La Honda road; but this experience is not without significance. At least it strengthens my contention that inherited tendency is less strong than environment, and that in some cases, at least, inherited tendency must be called into action by some specific stimulus or combination of stimuli operating upon the plant from outside itself. In our white redwoods, the descendants of an exceedingly ancient race of trees in which heredity should be proportionally strong, we have a certain amount of evidence that the irritability and the power of response of the organism to external influences are stronger than its heredity. May not this always be the case? May it not be that what we call heredity is really the response to similar stimuli and combinations of stimuli occurring in orderly succession in the course of nature?

STANFORD UNIVERSITY,
CALIFORNIA,
June, 1900.

BIBLIOGRAPHY.

1895. FRANK, A. B. Krankheiten der Pflanzen. Bd. I. Breslau.
1896. ——The same, Bd. III.
1899. GANNETT, HENRY. The Redwood Forests of the Pacific Coast. *Nat. Geograph. Magazine*, Vol. X.
1898. GOEBEL, KARL. Organographie der Pflanzen. Bd. I. Jena.
1893. PEIRCE, GEORGE J. On the Structure of the Haustoria of Some Phanerogamic Parasites. *Annals of Botany*, Vol. VII.
1864. SACHS, JULIUS VON. Ueber den Einfluss der Temperatur auf das Ergrünen der Blätter. *Flora*, 1864. Reprinted 1892, in *Gesamm. Abhandlungen*, Bd. I.
1894. TRELEASE, WILLIAM. Leitneria floridana. *Sixth Ann. Report Missouri Bot. Garden.*
1893. ZIMMERMANN, A. Botanical Microtechnique. Translated by J. E. Humphrey. New York.

EXPLANATION OF PLATE XIV.

All figures were drawn with an Abbé camera lucida.

Fig. 1. Cross-section of green redwood leaf, showing position of vascular bundle and resin-tubes, the well developed palisade parenchyma and sclerenchyma. × 30. Diagrammatic.

Fig. 2. Cross-section of a larger white redwood leaf, showing the absence of palisade parenchyma and sclerenchyma. × 30. Diagrammatic.

Fig. 3. Strip of epidermis from upper side of green redwood leaf, showing distribution of stomata near midrib. × 60.

Fig. 4. Similar strip from white redwood leaf, showing distribution of stomata in an equal area. × 60.

Figures 3 and 4 are from mountain redwoods.

Fig. 5. Similar strip from green redwood leaf, showing number and distribution of stomata. × 60.

Fig. 6. Similar strip from white redwood leaf, showing the same. × 60.

Figure 5 is from redwood growing in Arboretum of Stanford University, fig. 6 from mountain redwood.

Fig. 7. Part of green leaf shown in fig. 1, from above vascular bundle, showing palisade parenchyma, sclerenchyma, starch-grains, and other contents of palisade cells. × 300.

Fig. 8. Part of figure cut off by dotted line, showing starch-grains and other contents of mesophyll cells, thick-walled sclerenchyma and epidermis at edge of leaf, strengthening cells around resin-tube, etc. × 300.

Fig. 9. Corresponding part of fig. 2, a white redwood leaf, showing unorganized contents of mesophyll cells, lighter strengthening tissues, etc. × 300.

Fig. 10. Showing structure of stoma of white redwood and absence of chlorophyll and starch-grains. × 300.

Figs. 11 and 12. Two views from the surface of stoma from white redwood leaf; fig. 11 from above, showing auxiliary cells *(Nebenzellen)*; fig. 12 from further down, showing guard cells. Stoma closed. × 350.

Fig. 13. Surface view of epidermis and stoma from green redwood leaf, showing larger and more vigorous epidermal cells, and decidedly thicker walls of epidermal cells. × 350.

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A REVISION OF THE GENUS CALOCHORTUS.

BY CARL PURDY.

PLATES XV-XIX.

INTRODUCTION.

THE most widely diffused as well as the handsomest of the liliaceous plants of the Pacific Coast are the Calochorti. On the north they reach British America; one species is to be found as far east as Nebraska; several are natives of northern Mexico; and within these limits no considerable section of country is destitute of some species.

While the range of the genus is so immense, that of several of the species is also very extensive. What a diversity of conditions *C. nuttallii* meets in its range from the western side of the Sierra Nevada to Nebraska, and from the Snake River to Arizona. *C. nitidus* is found from the meadows of eastern Oregon to the shores of Yellowstone Lake, *C. albus* from San Diego to Tehama, and many others are scattered over hundreds of miles. A species distributed over a region so varied in soil, climate, and altitude cannot but be variable; and it hardly need be added that the genus *Calochortus* is a very difficult one for botanists to deal with.

I long since became convinced that it is only in the garden, where plants from different localities can be grown under identical conditions, that the relationship between apparently different forms can be satisfactorily determined. For some years I have grown a large variety of Calochorti in my grounds, and have had nearly every known species under cultivation, often in many forms. The culture of Calochorti is most interesting, though not unattended with cultural difficulties; but the beautiful flowers amply repay all efforts, and the garden has proved the identity of forms

apparently different; here, also, variations attributed to environment are shown to be constant. In the garden, too, strains, which from a botanist's standpoint seem scarcely distinguishable, show marked differences in vigor, flowering time, or immunity from disease.

It is a peculiarity of our liliaceous plants, that as a rule in a given locality there is little variation from a well marked type, as little, indeed, as may be found between flowers growing upon the same plant. Hundreds and thousands of flowers may be picked, all conforming closely to this type. In another locality, the same species will be found markedly different. The difference between the forms in the two localities may be slight, consisting merely of a marking or a slightly varying leaf, habit, or gland; yet the variant, once noted, is found to be constant. In *Calochortus* color forms are frequent, the flowers from one bulb retaining the same tints under any and all conditions. The difference between forms from different localities is rather that which florists designate by the word "strain" than what is usually understood to constitute a botanical species or variety.

In cultivation it has frequently been found that a very slight variability in strains is accompanied by a marked constitutional difference. In two beds of *Calochortus venustus*, planted in the same soil, and separated only by a thin board, it would puzzle a botanist to state wherein the plants vary. They come from widely separated localities, and the difference is one more easily detected by the eye than conveyed by words. In one bed, two-thirds of the leaves are already destroyed by mildew *(Botrytis)*, while in the other, not one leaf is injured; and such is the case whenever and wherever the two are planted. Many similar instances occur in other species, but a single one is sufficient to show that the slight variations which the eye detects are not the only ones.

Such strains are present in nearly every species of *Calochortus*. The range of a strain may be very local—a few miles square—or it may be found over half the length of a state. In *Calochortus venustus* one strain runs through all

the plants found for hundreds of miles along the Sierra; another strain is found in the same species occurring in the Coast Range and over an equal area. In some of the more variable species there are several strains.

In many of the Calochorti the gradations from one species to another are so slight that it is impossible to separate them. The extreme types on which the species are founded are easily distinguishable, but a perfect chain of variations links them closely together. There is no doubt that *C. weedii*, *C. plummeræ*, and *C. obispoënsis* are variations of a greater species.

While, as before stated, it is the rule that a given locality produces specimens conforming closely to a type, yet this is not always the case. In some localities the variations are bewilderingly numerous. I have seen places where hundreds of flowers of *C. venustus* could have been selected, each differing in color and markings from the rest. Why a species that remains so true to a type in some localities should vary so remarkably in others is a subject that will not be discussed at present. Hybridization will account for it in some instances, while in others it is hardly a tenable hypothesis.

I cannot say that I attribute any material share in the origin of the many strains or varieties to hybridization, although among the Calochorti it is not infrequent. Such crosses as *C. albus* and *C. benthami*, *C. maweanus* and *C. pulchellus* are frequently met with, but I have never yet seen one that was fertile. Again, varieties of a species, e. g., *C. luteus* var. *oculatus* and *C. luteus* var. *citrinus*, readily cross and produce fertile hybrids. Over a small area innumerable cross-breeds may be found, but a few miles away the two varieties will be found separate and varying as little as in any locality. Then again, hybridization often will not take place between two apparently very closely related species. I have often seen *C. vesta* in flower surrounded by large numbers of *C. luteus* var. *oculatus* or var. *citrinus*, but not a plant could be found that in any way indicated hybridization; while last summer, in the Sierra Nevada, I

visited a spot where *C. luteus* var. *oculatus* and *C. luteus* var. *citrinus* had hybridized to a remarkable degree. A few yards away, indeed mingling with them, were thousands of *C. venustus*. Within a quarter of a mile I do not doubt there were fifty thousand plants in flower, yet close search failed to reveal one that in any way suggested a cross of *C. venustus* with any of the variations of *C. luteus*.

In the "Botany of California," Vol. II, published in 1880, Watson described twenty-six species of *Calochortus*. The work was carefully done, though the material at command was meager as compared with that obtainable now; yet nearly every species recognized by Watson stands good to-day. Many new species have been added, but by the exploration of new territory rather than by the subdivision of old species.

The work of preparing the present paper has been facilitated by the courtesy of the California Academy of Sciences and the University of California, in allowing me to inspect their herbarium specimens. Mr. J. W. Congdon of Mariposa very courteously permitted an examination of his material, and to Mr. G. W. Hansen I am indebted for a set of specimens from Amador County. My personal collection, including both herbarium and living specimens, covers a wide range; still, with these facilities, probably as good as can be obtained anywhere, the material is painfully unsatisfactory in some species, several of which are represented in the best herbariums by a single specimen, if at all.

With each year appear many new forms, even from California. Last season brought three new species, and many striking variations of old species were added to the already large assortment. The field is immense and has never been properly worked over. In view of these facts, it seems the wisest course to disturb existing nomenclature as little as possible. As to whether a given degree of difference warrants a specific or a varietal name seems to me to be very largely a matter of personal opinion. While one can hardly agree with the author who designates a color form by a specific name, it will probably be consulting the convenience of

botanists to allow such a name to stand for the present, especially as a more extensive knowledge of the subject may result in still further changes. The pressing need is for a work containing descriptions of all known species of Calochorti, together with such grouping as will readily convey to the student the relationship of the various species.

In the "Botany of California," the types of the species known to Watson are usually very accurately described. The only criticism to be made is that in many instances he was acquainted with but a few representatives of the species. Nineteen years have added much to our knowledge of the range of the various forms of Calochorti, but it is still far from complete. In the notes on distribution of species, the range as I have accurate knowledge of it is given. A species may, and in many cases doubtlessly does, extend over a far wider range than that with which it is credited.

The measurements of any portion of a plant as given in published descriptions of Calochorti are of little value, and are apt to be misleading. Environment makes the greatest difference in the size of the plants. Take, for instance, those of the woods, such as *C. albus*, *C. pulchellus*, and the *elegans* group; the variations are almost limitless. Especially after a forest fire is growth luxuriant. Plants which under adverse conditions have leaves but a few inches in length, and few-flowered, slender stems, will, under more favorable circumstances, produce great leaves a foot or two long, stout stems eight inches to two feet in height, and a dozen or more fine flowers. Especially do the plants of the Mariposa section respond prolifically under fertile conditions. If the season be dry, the plants are sparsely scattered and but a few inches above ground; but let the season be one of great rainfall, they fairly hide the ground with tall, many flowered stems upholding numerous large blossoms. But while measurements based upon a series of specimens are almost valueless, proportional measurements of the parts of the same flower are often of great importance, the proportions between the parts being usually the same whatever the size of the flowers.

In the present paper I have departed from the usual method by describing a single type plant and indicating the variations in the notes. Personal experience has proven that the fuller a description is, the more value it has in the determination of specimens; details seemingly of little importance at the time when the specimen was described, are often indispensable in the work of later students, making it an absolute necessity for them to refer to the type specimen in order to determine what was really described. This brevity in some of the earlier descriptions makes them entirely valueless for determination.

Having no personal knowledge of the Mexican species outside of the herbarium, I have not included them in this revision, being unable to add anything of value. There is a large and almost unknown field in Arizona and western New Mexico which will probably yield several new species.

Only the description and when possible the locality (the original locality is quoted in nearly every instance) of the species have been given in the following pages, the synonymy having been omitted because of its often doubtful nature.

KEY TO THE SPECIES OF CALOCHORTUS.

SECTION I. *Eucalochortus.*

Flowers or fruit nodding; petals incurved or strongly arched; gland transversely crested or hairy; capsule nodding, with thin acute or winged cells; leaves long and glossy, not channeled.

Group 1. GLOBE TULIPS.

Type of Group *C. albus.*

Flowers subglobose, nodding. Woodland plants; California.

Flowers white; petals covered with scattered silky hairs within. 1. *C. albus.*

Flowers rose color; petals silky within, partly opening out. Foothills of Fresno and Tulare counties, California..2. *C. amœnus.*

Flowers light yellow; petals silky within, gland bordered with stiff hairs which cross each other.....3. *C. pulchellus.*

Petals very strongly inarched, not silky within, but margin thickly set with short, stiff hairs; gland like last.....4. *C. amabilis.*

Group 2. STAR TULIPS.

Type of Group *C. elegans.*

Flowers campanulate, erect or ascending; capsule nodding (except in No. 13); stem low and flexuous (In 14, 15, 16, stout and erect), not bulbiferous or very seldom so.

**Petals covered with hairs, and with a transverse scale covering upper part of the gland. Woodland plants.*

Flowers yellow. Foothills of the Sierra Nevada...5. *C. benthami.*
Flowers white or purplish blue, covered with long erect hairs; capsule oblong-elliptical; stem branching.....6. *C. maweanus.*
6*a*. *C. maweanus* var. *major.*
6*b*. *C. maweanus* var. *roseus.*
Flowers blue, covered with silky hairs, longer and slenderer than the last; capsule orbicular; inflorescence umbellate.
7. *C. cæruleus.*
Flowers greenish white; petals with very narrow scale and covered with long hairs. Oregon and north..8. *C. elegans.*
8*a*. *C. elegans* var. *nanus.*
Flowers yellow, green tinged; petals strongly inarched and pit deeply set. Mt. Jefferson, Oregon.....9. *C. lobbii.*

***Petals with a transverse scale closely appressed over upper portion of gland, nude or nearly so. Woodland plants; in dry soil.*

Flowers white with a single tuft of a few hairs at each end of scale on petals; plants very low and slender. Sierra Nevada.
10. *C. nudus.*
Flowers white with scant hairs on lower third; plants taller than the last. Vicinity of San Francisco Bay.....11. *C. umbellatus.*

****Petals nude or only lower portion hairy; flowers campanulate; plants growing in open wet meadows.*

Flowers lilac, hairy on lower third; one or several bulblets on stem below the ground.....12. *C. uniflorus.*
Flowers white, not bulbiferous; capsules erect...13. *C. shastensis.*

*****Petals covered with silky hairs; flowers and stems stout and erect. Closely related to Group 1 of Mariposa. Plants growing in open fields or hillsides. Mt. Shasta, California, and north.*

Flowers blue; petals without scale, covered with long silky hairs. Mt. Shasta, California (?), and Willamette Valley, Oregon.
14. *C. tolmiei.*
Flowers white; petals with scale, otherwise the same as the last. Willamette Valley, Oregon.....15. *C. purdyi.*
Flowers straw color; petals without scale, otherwise like *C. tolmiei.* Lake Pend d. Oreille, Idaho.....16. *C. apiculatus.*

SECTION II. *Mariposa.*

Flowers open-campanulate; gland usually densely hairy; capsule and pedicels erect.

Group 1. OREGON MARIPOSAS.

Type of Group *C. nitidus.*

Capsule as in Section I, but erect; leaf (as in Section I) long and glossy, not channeled. Oregon and northeast.

**Petals with an indigo blotch in the center.*

Flowers large, white to lavender .....17. *C. nitidus.*

***Petals not spotted in the middle; flowers lilac, smaller than in the preceding.*

Flowers strongly arched and barred with yellow.....18. *C. greenei.*
Flowers less arched; stem bracted midway.....19. *C. pavonaceus.*
Similar to last; stem not often bracted midway. 20. *C. longebarbatus.*
Flowers white, densely hairy above gland.....21. *C. howellii.*

Group 2. ROCKY MOUNTAIN MARIPOSA.

Type of Group *C. gunnisoni.*

Gland transverse and narrow; leaf usually as in Section I. East of the Rocky Mountains..... .....22. *C. gunnisoni.*

Group 3. WEED'S MARIPOSA.

Type of Group *C. weedii.*

Petals covered with slender hairs; capsule narrowly oblong, with thick, obtusely angled cells; radical leaf as in Section I, solitary, long, shining, and not channeled; bulb heavily coated with coarse black fiber.

Flowers orange or rarely pink or white.....23. *C. weedii.*
Flowers purple...23*a*. *C. weedii* var. *purpurascens (C. plummeræ).*
Petals brownish, short, truncate, not equaling sepals.
23*b*. *C. weedii* var. *vestus.*
23*c*. *C. weedii* var. *obispoënsis.*

Group 4. GOLDEN BOWL MARIPOSAS.

Type of Group *C. clavatus.*

Petals yellow, lower half covered with clavate hairs; radical leaves linear and deeply channeled.

Stem stiffly zigzag.....24. *C. clavatus.*
Stem not zigzag. Doubtfully placed in this group, but has no clavate hairs...25. *C. concolor*, sp. nov. *(C. luteus* var. *concolor).*

Group 5.

Type of Group *C. kennedyi.*

Petals nearly naked; gland round, small, and densely hairy with matted hairs; leaves ashy blue, linear, deeply channeled. Desert plants.

Petals vermilion or orange.....26. *C. kennedyi.*
Flowers clear yellow; petals densely hairy below; capsule narrowly oblong.....27. *C. aureus.*

Group 6. BUTTERFLY TULIPS.

Type of Group *C. venustus.*

Petals slightly hairy below, usually oculated and brilliantly colored; gland prominent, round or lunate; leaves linear, channeled. California.

Flowers yellow; petals not oculated; gland lunate; capsule attenuate from a broad base; plant dwarfed.....28. *C. luteus.*
Flowers yellow or lemon, otherwise same as in var. *oculatus.*
28*a*. *C. luteus* var. *citrinus.*
Flowers white, yellow or lilac; petals oculated, gland lunate.
28*b*. *C. luteus* var. *oculatus.*
28*c*. *C. luteus* var. *robusta.*
Flowers lilac or white; gland narrow, doubly lunate...29. *C. vesta.*
Flowers white, cream, lilac, purple, red or pink; petals oculated, in some varieties with a red blotch above eye; gland round; capsule linear. .....30. *C. venustus.*
30*a*. *C. venustus* var. *roseus.*
30*b*. *C. venustus* var. *eldorado.*
30*c*. *C. venustus* var. *purpurascens.*
30*d*. *C. venustus* var. *sulphureus.*

Group 7. LILAC MARIPOSAS.

Type of Group *C. splendens.*

Petals white, lilac, or purplish, not oculated, more or less hairy; gland small, round, and densely hairy; leaves linear, channeled.

Flowers lilac-purple; petals hairy on lower third. 31. *C. splendens.*
Flowers lilac to salmon, short yellow hairs on lower third of petal; plants low and slender.....31*a*. *C. splendens* var. *montanus.*
Flowers large; petals pale lilac with cobwebby hairs on middle third... .....31*b*. *C. splendens* var. *major.*
Much larger and stronger than type; hairy on lower third of petal.
31*c*. *C. splendens* var. *rubra.*
Flowers white; gland ill defined; plants more slender than last. Colorado Desert.....32. *C. palmeri.*
Flowers white or lilac with dark maroon spot at base of petal; capsule oblong. Resembles *C. splendens*.....33. *C. catalinæ.*
Flowers smoky white; stems stout and umbellate.
34. *C. invenustus.*
Flowers similar to last; petals deeply pitted.....35. *C. excavatus.*

Flowers purple; sepals obtuse; stem flexuous and weak, almost creeping.....36. *C. flexuosus.*
Flowers white. Resembles *C. splendens*.....37. *C. dunnii.*

Group 8. GREEN BANDED MARIPOSA.

Type of Group *C. macrocarpus.*

Petals purplish lilac, with a greenish line down the back, obovate-acuminate.

Stems stout and rigid; leaves linear and deeply channeled. Northeastern California to eastern Washington and southern Idaho. 38. *C. macrocarpus.*

Group 9. SEGO LILIES.

Type of Group *C. nuttallii.*

Petals white, lilac, yellow, or pink; gland round; stem prominently bulbiferous at base, umbellate.

Flowers as described above. Stout desert plants of the Great Basin and eastwardly.....39. *C. nuttallii.*
Flowers smaller, usually white; anthers sagittate. Slender Alpine plants. Sierra Nevada.....40. *C. leichtlinii.*

DESCRIPTION OF SPECIES.

Calochortus.

Perianth deciduous, of six distinct, more or less concave segments, the three outer (sepals) greenish and more or less sepaloid, the inner (petals) mostly broadly cuneate-obovate, usually with a conspicuous glandular pit near the base, and variously colored. Stamens six, on the base of the segments, included; anthers linear to oblong, basifixed, dehiscent laterally. Ovary sessile, triquetrous and three-celled, many ovuled; stigmas sessile, recurved, persistent; capsule elliptical to lanceolate, membranous, three-angled or three-winged, mostly septicidally dehiscent; seeds numerous, two rows in each cell, somewhat flattened, with a thin membranous white or brownish, often loose, testa. Stems usually flexuous and branching from membranous or but fibrous-coated corms; leaves few, linear-lanceolate, radical and cauline, the latter alternate and clasping; all with many nerves and transverse veinlets. Flowers one to twenty, showy, terminal, paniculate or umbellate.

The above generic description is in greater part that of Sereno Watson as given in the "Botany of California."

The genus is confined to western America, from Nebraska to the Pacific Ocean, and from northern Mexico to British America.

SECTION I. *Eucalochortus.*

Flowers or fruit more or less nodding; petals strongly incurved or arched, with a broad, transversely crested or more or less hairy pit above the base;

sepals naked, rarely spotted; capsule elliptical or broadly oblong, deeply triquetrous and septicidal, the thin compressed lobes acute or winged; seeds ascending, close and pitted, the testa mostly brownish.

Group 1. GLOBE TULIPS.

Flowers subglobose, nodding. Woodland plants; California.

1. Calochortus albus *Dougl.*

Calochortus albus DOUGLAS *in litt.*

Cyclobothra alba BENTHAM, Trans. Hort. Soc., N. S., Vol. I, p. 413, Pl. XIV, fig. 3; Bot. Register, Vol. XX, 1835, Tab. 1661.

Stem stout, glaucous, usually branching, a foot or two high; radical leaves a foot or two long, 8–12 lines wide, lanceolate-acuminate; bracts large and foliaceous, 1–5 inches long, 4–8 lines wide; sepals shorter than petals, ovate-acuminate, greenish white; petals pure white, purplish at base, ovate-orbicular, acutish, 12–15 lines long, with scattering long silky hairs above the gland; gland lunate, shallow, with four transverse imbricated scales, fringed with close short yellow or white glandular hairs; anthers oblong-obtuse, mucronate; ovary attenuate above; capsule 1 or 2 inches long, 6–12 lines broad, abruptly short-beaked; seeds brown, pitted.

The original specimens are in all probability from Monterey, as Douglas visited there, where the species is plentiful.

C. albus is found in the Coast Range of California, from San Francisco Bay south to San Diego County, and in the Sierra Nevada Mountains, from Butte County south to San Diego County.

There is quite a difference between the plants of the Coast Range and those of the Sierra Nevada. The flowers of the former are darker in color, often tinged with rose, and with petals thicker, more strongly inarched. The petals never open out sufficiently to show the inside of the flower, which after being in bloom a few days is half opened. *Calochortus amœnus* Greene is really a color form of this Sierran form of *Calochortus albus;* but owing to the fact that the original locality of *C. albus* is unknown, the writer hesitates to erect either the form from the Coast Range or that from the Sierra Nevada into a new species.

Several variations of the form of the Coast Range have been described, some as species and some as varieties; but I fail to discover any characters by which they may readily be recognized.

2. Calochortus amœnus *Greene.*

Calochortus amœnus GREENE, Pittonia, Vol. II, 1890, p. 71.

Similar to *C. albus*, but lower and more slender; petals a rosy pink with the gland rose-purple, scarcely at all arched, and opening in full bloom.

Found in the higher foot-hill region of Fresno and Tulare counties, California.

"Mountains east of Visalia, California."

3. Calochortus pulchellus *Dougl.*

Calochortus pulchellus DOUGLAS *in litt.*
Cyclobothra pulchella BENTHAM, Trans. Hort. Soc., N. S., Vol. I, 1835, p. 415, Pl. XIV, fig. 1; Bot. Register, Vol. XX, 1835, Tab. 1662.

Stem stout, glaucous, usually branching, 8 to 16 inches high; radical leaves a foot long, 6 to 12 lines wide, lanceolate-acuminate; bracts large and foliaceous, 2 to 3 inches long on the same plant, 4 to 6 lines wide; sepals shorter than petals, ovate-acuminate, yellow tinged with brown on the back; petals canary-yellow, ovate with the base cuneate, obtuse at apex, 9–12 lines long, with scattering long silky hairs above the gland, and bordered with short stiff hairs; gland deep, protruding outwardly, bordered with stiff hairs which cross each other; anthers oblong-obtuse; ovary elliptical to elliptical-orbicular, abruptly short-beaked.

The original specimens of this species were collected by Douglas prior to 1835. The exact locality is not given; but the only place in which the species has since been found is in the Mount Diablo region, a section which was easily accessible at the time of Douglas' visit to California, and often visited by his Mexican-Californian hosts.

Although in a region much visited by botanists since then, no specimens were to be found in any of the herbariums of this State up to the year 1897, when *C. pulchellus* was collected by Miss Alice Eastwood of the California Academy of Sciences.

The very name *Calochortus pulchellus* had been appropriated by another yellow-flowered species of the same group, which is described and named below as *C. amabilis*, and which is clearly a distinct species.

Following are the original descriptions of the species copied from the Botanical Register:—

Cyclobothra pulchella.—"Umbellis 2-3 floris, pedunculis bracteis brevioribus, floribus globosis, petalis ovatis obtusis serrulato-fimbriatis foveâ valdé excavatâ extus callosâ, sepalis ovato-lanceolatis acuminatis vix brevioribus."

Calochortus pulchellus.—"Caulis erectus, teres, glaber, subcorymbosus, apice magis ramosus quam in præcedente, et humilior. Folia plana, acuminata, minus glauca; superioribus brevioribus. Pedunculi bracteis foliaceis breviores, bini ternive. Flores globosi, minores quam in præcedente, lutei. Sepala virescentia, viridi-striata, petalis paululum breviora, acutissima. Petala ovata, barbata, fimbriata, basi glabra: foveâ nectariferâ pilis absconditâ."

4. **Calochortus amabilis**, sp. nov.

Stems stout, usually branching in pairs, 8 to 12 inches high, glaucous; radical leaves 10 inches long, 4 to 6 lines wide, lanceolate-acuminate, tinged with purple; bracts large and foliaceous, 2 to 3 inches long, 4 to 6 lines wide; sepals shorter than petals, ovate, shortly cuneate at base, sharply acuminate or even mucronate at apex, yellow tinged with brown on the back; petals clear yellow, ovate, with a short claw, obtuse at apex, naked but margined with a close row of short stiff hairs, very strongly inarched so that the tips of the petals overlap each other much like a child's pin-wheel; gland very deep, projecting upwards and outwards like a knob, lined with short stiff hairs which cross each other; anthers oblong-obtuse; ovary elliptical, short-beaked.

C. amabilis is found on the hills along the north side of San Francisco Bay, from the redwood belt to the Sacramento foot-hills, as far north as Burnt Ranch, Trinity County, California.

The species has been distributed in large numbers among the flower-growers of the world as *Calochortus pulchellus*, which it resembles in habit and size. The latter is more closely allied to *C. albus*, having the same globular flower and petals silky-haired within. It is also of a much lighter shade of yellow, and never could be confused with *C. amabilis* by anyone who had seen both.

Group 2. STAR TULIPS.

Flowers campanulate, erect or ascending; capsule nodding (except in No. 13); stem low and flexuous (in 14, 15, 16, stout and erect); not bulbiferous or very seldom so.

* *Petals covered with hairs, and with a transverse scale covering upper part of the gland. Woodland plants.*

5. Calochortus benthami *Baker*.

Calochortus benthami BAKER, Journ. Linn. Soc. (Bot.), Vol. XIV, 1875, p. 304; also Bot. Mag., Pl. 6475.

Stems several-flowered, very flexuous, branching, dividing into pairs 7 inches high; radical leaf linear-lanceolate, shining, 4 lines wide, exceeding stem; bracts ovate-lanceolate, long, acuminate, 2 to 4 inches long, 3 to 5 lines wide; sepals 8 to 10 lines long, narrowly ovate, mucronate; petals yellow, with a yellow claw, naked, obovate, rounded above, a little longer than sepals, the upper portion of the gland covered by a narrow crescentic scale which is densely bordered above with short yellow hairs, some of which are clavate; anthers lanceolate-acute, capsule nodding, nearly orbicular, 6 to 9 lines long.

Found in the lower Yellow-Pine belt of the Sierra Nevada, from Mariposa to Butte counties.

"California."

The description is drawn from rather an extreme specimen. The plants are often very slender, simple, and but three to four inches in height. The flowers vary but little. In some sections flowers with the claw dark red or nearly black are common (*C. wallaceii* Hort.).

6. Calochortus maweanus *Leichtlin*.

Calochortus maweanus LEICHTL. ex BAKER, in Journ. Linn. Soc. (Bot.), Vol. XIV, 1875, p. 305; also Bot. Mag., Pl. 5976.

Resembles the preceding; stem usually branching, very slender and flexuous, 3–10 inches high, with from a few to ten flowers; leaves glaucous, much exceeding stem, 2–6 lines wide; bracts lanceolate, narrow, 10 lines or more long; sepals ovate-lanceolate, acute or acuminate; petals a little longer, broadly ovate, acute; gland covered above with a narrow transverse scale, immediately above the scale densely hairy, the entire surface thickly bearded with long, erect, white or bluish hairs; anthers lanceolate-acuminate, 2–3 lines long; capsule oblong-elliptic.

This is the type which is found in the Coast Range, from San Francisco Bay, at least as far north as Trinity County, California, and western Oregon.

"California."

6*a*. *C. maweanus* var. *major*, var. nov. This variety is twice as large as the type, from which it differs in its much stronger habit and lighter color.

Grows in the Yellow-Pine belt, Butte County, California.

6*b*. *C. maweanus* var. *roseus*, var. nov. The flowers of var. *roseus* are tinged with rose; the bulb is distinctive, having a smooth, mahogany-colored coat.

Its habitat is western Oregon.

7. Calochortus cæruleus *Watson*.

Calochortus cæruleus WATSON, Proc. Amer. Acad., Vol. XIV, 1879, p. 263.

In general resembling a small specimen of *C. maweanus*. The plants are very slender; leaves and bracts narrower, pedicels more slender; flowers almost always in an umbel, petals more rhombic in outline, claw more slender, scale broader and fringed, the remainder of petal densely covered with long slender silky hairs; anthers oblong-obtuse; capsule orbicular, not beaked, 6 lines long.

Specimens of this species show but little variation.

"California (in the Sierra Nevada, Placer to Plumas counties)."

8. Calochortus elegans *Pursh*.

Calochortus elegans PURSH, Fl. Am. Sept., Vol. I., 1816, p. 240.

Scape very slender, 4-8 inches high; leaves lanceolate-acuminate, narrow, exceeding scape; flowers one to four in umbel, bracts one-half length of pedicels, acuminate from a base 2 lines wide; sepals ovate-acute, greenish white without, lighter within, purplish at base; sepals obovate-obtuse, whitish or tinged slightly with green, with purple spot on claw, covered thickly with rather short soft hairs, which are white on upper and purple on lower portion, excepting that the margin and a band around upper portion of petal is naked; scale narrow, ascending, and deeply fringed, covering about one-third the width of claw; anthers long, acuminate; capsule elliptical, rounded at each end.

The type specimens were collected by the Lewis and Clark expedition on the headwaters of the Kooskoosky in Idaho (?).

The writer was never able to obtain specimens of this species until just as the present paper was going to press, when flowers which are unquestionably the true *C. elegans* were received from a collector in western Idaho, near Spokane. As the original description of the species is very brief, the fuller description, as given above, was drawn

from these specimens. There is much difference in general appearance between the species and var. *nanus*, and in my opinion the variety will eventually follow the other forms which were originally included under *C. elegans*, and be given specific rank. For the present, however, it is left under the varietal name. No more definite locality than western Idaho can be given for the type.

Some Californian specimens have been referred to *C. elegans* but the writer has never seen any which come under the species and does not believe that either the type or any of its variations are found in California. The Californian plants referred to as *C. elegans* are *C. nudus*.

8a. *C. elegans* var. *nanus* WOOD. (Proc. Phil. Acad., 1868, p. 168.) Dwarf and very slender, leaves very narrow; petals more hairy and ciliate, often acute or even acuminate.—*C. lyallii* Baker, Journ. Linn. Soc. (Bot.), Vol. XIV, 1875, p. 305.

"High hills, Yreka [California]. Also on Mt. Hood [Oregon]."

The description is that of Watson in the Botany of California.

The variety is known to the writer as it grows on Mt. Hood and on Mt. Adams. Watson evidently had small specimens, as under favorable conditions it is quite as strong as any of the preceding. The color is a delicate cream.

9. Calochortus lobbii, sp. nov.

Calochortus elegans var. *lobbii* BAKER, Journ. Linn. Soc. (Bot.), Vol. XIV, 1875, p. 305.

Stem 3 to 5 inches high, not so slender as the preceding; leaf a little exceeding stem in length, 3 to 5 lines wide, lanceolate, abruptly acute; sepals ovate-lanceolate, acute, greenish with a dark spot below, 6 to 8 lines long; petals a little longer, white tinged with green, broadly rhombic-ovate, very deeply pitted, the pit showing as a prominent knob on back of petal; scale very narrow, deeply bordered with long, feathered, hairy fringes, and concealed in the recess of pit; above the scale there is a nectar-producing gland covered by a broad band of agglutinated feathered hairs, above this band lower half of petal densely hairy with silky hairs, upper half less densely hairy; filaments subulate; anthers oblong-acuminate, ending in a hook-like cusp; capsule narrowly beaked.

So far found only on Mt. Jefferson, Oregon.

"Oregon."

The above was identified for the writer by Baker. As the other varieties which Baker mentions in his "Tulipeæ" have been erected into species, and this is more distinct in character than any of them, it seems proper to raise it to specific rank.

***Petals with a transverse scale closely appressed over upper portion of gland, nude or nearly so. Woodland plants; in dry soil.*

10. Calochortus nudus *Watson.*

Calochortus nudus WATSON, Proc. Amer. Acad., Vol. XIV, 1879, p. 263.
Calochortus elegans var. *subclavatus* BAKER, Journ. Linn. Soc. (Bot.), Vol. XIV, 1875, p. 305.

Low and slender, scape 2 to 4 inches high, with a single leaf 3 to 6 inches long, 3 to 6 lines wide, light green, of even width for most of length, abruptly acute; flowers one or more, in all specimens examined in an umbel if more than one; sepals narrowly oblong-ovate, acute, shorter than petals; petals greenish white or lilac, greenish at base, obovate, somewhat acute, denticulate above, 5 to 7 lines long, the same in width, entirely nude except for a tuft of two or three short stiff hairs at each extremity of the narrow, closely appressed scale which covers the upper margin of gland; anthers blue, oblong, two-thirds the length of the subulate filaments.

Probably *C. elegans* var. *subclavatus* of Baker.

On north sides of high mountains in the pine forests of the Sierra Nevada, from Tulare to Plumas counties; in loose dry soils.

"California (in the Sierra Nevada, Yosemite Valley to Plumas County)."

The type as described from Tulare County is white, but there seem to be variations tending to lilac, and in some sections a nude petal. This is the smallest flowered of all the Calochorti.

11. Calochortus umbellatus *Wood.*

Calochortus umbellatus WOOD, Proc. Acad. Nat. Sci. Phila., 1868, p. 168.
Cyclobothra elegans var. TORR. ?, Pac. R. R. Rept., 1856, Vol. IV, p. 146.
Calochortus collinus LEMMON, Erytheæ, Vol. III, 1895, p. 49.

Stem low and branching, 3-15 inches high, flexuous; radical leaf exceeding stem, narrow, 3-4 lines wide; bracts foliaceous, acuminate, 1-4 lines

long, 3–4 lines broad at base; flowers five to ten; sepals greenish white, ovate-lanceolate, acute; petals white, broadly fan-shaped, denticulated, obtuse; scale triangular, ascending, appressed on upper portion of gland, which has many short white hairs just above it, remainder of petal nude; filaments slender; anthers 1 line long, oblong-obtuse; capsule short, oblong, 6 lines wide, 8 lines long.

A native of California, found on the lower mountains and hills back of Oakland, Berkeley, and Mills Seminary, and on Mt. Tamalpais.

"Oakland, California."

The species was long confused with *C. uniflorus*, and was described as *C. collinus* by J. G. Lemmon. Professor E. L. Greene called the writer's attention to the earlier description of Wood.

C. umbellatus can be confused only with *C. uniflorus*, from which its lack of bulblets and the situation in which it grows, as well as its color, easily distinguish it. It varies but little.

****Petals nude or only lower portion hairy; flowers campanulate; plants growing in open wet meadows.*

12. Calochortus uniflorus *Hook. & Arn.*

Calochortus uniflorus HOOK. & ARN., Bot. Beech. Voy. Suppl., 1841, p. 398, Tab. XCIV.

Stem low, flexuous, but often stout, usually branched, 4–8 inches high, with one to four bulblets below the surface; radical leaves broad, 4–6 lines wide, exceeding stem; bracts linear-lanceolate, long and conspicuous; flowers four to ten, in one to three umbels, on long flexuous pedicels 3–10 inches long; sepals ovate, lanceolate-acuminate, greenish lilac; petals cuneate, somewhat truncate, denticulate, 10–12 lines long, color lilac, often with a purple spot on each side of the scale, naked above, sparingly hairy immediately above the gland; gland shallow, not pitted, a narrow triangular scale appressed upward over upper center; filaments slender; anthers obovate-obtuse, lilac, 2 lines long, one-half length of filaments; capsule elliptical.

Except in the spots on petal or sepal there are few color variations.

From Monterey, California, northward in the Coast Range to Grant's Pass, Oregon. Found in wet meadows.

C. uniflorus was originally described by Hooker and Arnott from specimens collected on the Beechy Expedition. The specimens were without doubt collected either in the

vicinity of San Francisco or at Monterey. A good cut accompanies the description.

C. lilacinus was first described by Kellogg from specimens collected near Calistoga, Napa County, California. Dr. Kellogg's original description, together with a water color drawing, are in the Herbarium of the California Academy of Sciences.

After carefully comparing all the data contained in the descriptions of *C. uniflorus* and *C. lilacinus* with both fresh and herbarium specimens from Monterey, Calistoga, and a number of other localities, the writer is thoroughly convinced that the two species are the same. The description of *C. uniflorus* was published much earlier than that of *C. lilacinus;* it is drawn from a fair specimen of the species.

It is obvious that Dr. Watson confused *C. lilacinus* with the quite different *C. umbellatus.*

The writer has seen *C. uniflorus* in meadows high in the mountains where the plants grew very low and slender and were only one- to three-flowered. In rich ground several bulblets are produced annually, and if left undisturbed large and dense masses are formed, sometimes hundreds of them to the square foot. In soft ground the stems are apt to run along close to the surface a few inches to a foot before coming through, and in these situations plants a foot high with pedicels ten inches long are not uncommon.

13. **Calochortus shastensis,** sp. nov.

Scape low, slender, 4 to 10 inches high, but unusually erect, with a single shining light green radical leaf 3 to 6 inches long, of almost uniform width (3 to 6 lines), but abruptly acute at apex; bracts lanceolate, 6 lines long; sepals long, ovate, acute and acuminate, greenish without, lighter within, purple spotted near base; petals white or lilac, broadly fan-shaped, somewhat truncated above, denticulate, naked except that some few specimens have a few hairs above the narrow, fringed, ascending scale which divides the gland; anthers linear, obtuse, slightly sagittate; capsule as in preceding but erect.

Found in open moist meadows in the vicinity of Sissons, California, at the base of Mt. Shasta, and about springy places on the western flank of the mountain.

C. shastensis has long been known and collected as *C. nudus,* which it closely resembles in flower but from which

it is clearly distinguished by the erect capsule. It is a curious fact that a species linking the small Calochorti of the wet lands with the *C. nitidus* section should be found at the very point where the latter terminates its most southern extension.

The true *C. nudus*, it will be noted, grows only on dry slopes in the Sierra Nevada, from Plumas County, California, southward.

GIANT STAR TULIPS.

****Petals covered with silky hairs; flowers and stem stout and erect. Closely related to Group 1 of Mariposa. Plants growing in open fields or hill-sides. Mt. Shasta, California, and north.*

14. Calochortus tolmiei *Hook. & Arn.*

Calochortus tolmiei HOOK. & ARN., Bot. Beech. Voy. Suppl., 1841, p. 398.

Stem stout, erect, usually branched, 9 to 18 inches high; leaves 4 to 6 lines broad, not greatly exceeding the stem, bracts foliaceous; petals 9 to 15 lines long, very broadly obovate and scarcely acute, rather deeply pitted, covered and fringed with long purple and white hairs; gland without scale, but the upper circular edge with a dense fringe of reflexed hairs; anthers lanceolate and acuminate, 2 to 3 lines long; capsule broadly elliptical, acutish at each end, 10 to 15 lines long.

C. tolmiei is frequently confused with *C. purdyi*. The writer has seen specimens of the true *C. tolmiei* from the hills west of the Willamette River in Oregon. Dr. Watson also refers specimens from Mt. Shasta, California, and Mt. Adams, Washington, to this species. The original locality is "Banks of the Walamet River."

15. Calochortus purdyi *Eastwood.*

PLATE XV.

Calochortus purdyi EASTWOOD, Proc. Cal. Acad. Sci., 3d Ser. (Bot.), Vol. I, p. 137, Pl. XI, figs. 8*a*–8*f*.

Glabrous and glaucous; stem 2 to 3 dm. [18 to 16 inches] high, rather stout, erect, branching, two to many-flowered, not bulbiferous at base; radical leaf solitary, sheathing the stem, linear-lanceolate, acuminate, 2 dm. [8 inches] long, 1 cm. [6 lines] wide, the upper surface bright green, the lower glaucous and ribbed with the filiform nerves; bracts foliaceous, lanceolate-acuminate, amplexicaul, upper ones opposite; pedicels equalling or slightly surpassing

the bracts, erect in flower, recurved in fruit; flowers broadly open-campanulate; sepals from elliptical to narrowly ovate, abruptly acuminate, tinged with purple on the outer surface, purple-veined on the inner, two-thirds as long as the petals; petals broadly obovate-cuneate, acute or rounded at apex, creamy white or tinged with purple, bearded all over the inner surface with long hairs which are white on the upper half of the petals, purple on the lower, somewhat arched by the narrow, transverse, semicircular, conspicuous gland, the shallow pit of which is covered by a densely hairy narrow scale; anthers lanceolate, abruptly acuminate, cream color or purplish, shorter than the filaments, which broaden to the base; capsule 3 cm. [one-fourth inch] long, 2 cm. [10 lines] wide, broadly elliptical, with the three wing-like valves transversely veined.

The above is the original description as given by Miss Eastwood.

"Willamette Valley [Oregon], in the foot-hills."

C. purdyi may be distinguished from *C. tolmiei* by its color and the absence of the gland. The two are frequently confused and many of the specimens in herbariums labeled *C. tolmiei* are in reality *C. purdyi*.

Through *C. howellii*, *C. tolmiei* and *C. purdyi* are closely related to *C. longebarbatus;* while the large forms of *C. maweanus* link them to the *elegans* group.

16. Calochortus apiculatus *Baker.*

Calochortus apiculatus BAKER, Journ. Linn. Soc. (Bot.), Vol. XIV, 1875, p. 305.

Stem stout, erect, a foot to 18 inches high; the single radical leaf 6–12 inches long, 3–9 lines wide; bracts linear, acuminate, 1–3 inches long; sepals lanceolate, greenish white, acute, 6–9 lines long; petals straw colored, broadly obovate, one inch long, distinctly pitted, above with only scattering hairs, pit densely hairy and without scale; anthers 4 lines long, acuminate; filaments as long; capsule 12–15 lines long, narrowly oblong.

"Columbia brittanica ad montes Pend Oreille et Kootenay."

The above is drawn from Baker's original description and from letters. The writer has no personal knowledge of the species.

SECTION II. *Mariposa.*

Flowers and fruit erect on stout pedicels; flowers open-campanulate; gland usually densely hairy; capsule (except in Group 1) narrow, with thick lobes,

septicidal; sepals often hairy or subglandular, or spotted within; seeds ascending and somewhat turgid, with white, loose and spongy, minutely tesselated testa.

Group 1. OREGON MARIPOSAS.

Capsule with thin, acute or winged cells (as in Section I, but erect), also resembling Section I in the solitary, flat, shining leaf. Closely linked to the preceding group of Eucalochorti (Giant Star Tulips). Oregon and to the northeast.

**Petals with an indigo blotch in the center.*

17. Calochortus nitidus *Dougl.*

Calochortus nitidus DOUGLAS, Trans. Hort. Soc., Vol. VII, 1830, p. 277, Tab. IX, fig. *a*.

Stem bulbiferous near the base, slender, stiffly erect, not bracted in the middle, bearing an umbel of 2–4 flowers subtended by 2–4 linear bracts; pedicels often 3 inches long; sepals ovate-lanceolate, long-acuminate, exceeding petals; petals white to lavender, a conspicuous indigo spot in the middle, 2 inches long, the same in width, broadly cuneate, rounded above, with a small rounded gland densely matted with short hairs, and scattering long hairs around and above; filaments filiform, winged below; anthers linear-oblong, two-thirds as long as filaments; capsule elliptical-orbicular, strongly winged and crested.

Described from specimens from Union, Oregon.

"Columbia britannica" (Dougl.); "Oregon" (Spaulding).

This species is the largest flowered of the group; it is found in wet fields, from eastern Oregon through Idaho to Montana and northeastern Nevada. Specimens from Yellowstone Lake are yellow. The purple blotch on the petal seems to be distinctive.

***Petals not spotted in the middle; flowers lilac, smaller than in the preceding.*

18. Calochortus greenei *Watson.*

Calochortus greenei WATSON, Proc. Amer. Acad., Vol. XIV, 1879, p. 264.

Stem stout, branching, often a foot high or more, 2–5-flowered; leaf about equalling the stem, an inch broad; bracts narrow, elongated; sepals greenish with more or less of lilac within, and with a yellowish hairy spot above the base; petals broadly fan-shaped and obtuse, 1¼ to 1½ inches long, lilac, somewhat barred with yellow below, strongly pitted and arched, the lower part densely covered with very long yellow hairs, upper part of the blade more thinly hairy, not ciliate; pit densely villous above a broad transverse, laciniate scale; anthers broad, acute or obtuse, ½ inch long; capsule an inch long, 4 to 6 lines broad, attenuate into a stout beak.

"Siskiyou County, California (Greene); Multnomah County, Oregon (Howell)."

The description and locality are copied from the original of Watson, but the latter habitat is incorrect, as Howell's specimens are *C. longebarbatus.*

While at work on this revision the writer had not access to specimens of authentic *C. greenei*, the examples from Oregon and Washington referred to this species being in reality *C. longebarbatus* Watson. Specimens from Modoc County, California, a little east of the type locality, appear, however, to be the true *C. greenei.*

Professor Greene, the discoverer of the species, is of the opinion, which the writer also shares, that Dr. Watson confused two distinct species in his description. *C. greenei* is described as having the erect capsule characteristic of the section, while Professor Greene reported that some of his specimens had the nodding capsule of the Eucalochorti. From the evidence, it would seem that the densely hairy, strongly arched *Calochortus* described by Dr. Watson is properly another one of the Giant Star Tulips; but this cannot be definitely determined until there is fuller material to work with.

19. Calochortus pavonaceus *Fernald.*

Calochortus pavonaceus FERNALD, Bot. Gaz., Vol. XIX, 1894, p. 335.

Stem erect, stiff, slender, a foot or so high, with a single linear bract near the middle and two or more below the umbel; radical leaf flat, shining, lanceolate, not channeled; flowers 1–4 in an umbel; pedicels 1½–3 inches long, exceeding bracts; sepals purplish, ovate-lanceolate, acuminate; petals 1½ inches long, cuneate-obovate, denticulate, with a broad claw, color lavender to purple, with a circular band above the small round gland, which is covered with densely matted yellow hairs; a few hairs on the margin and silky hairs sparingly scattered over lower third; filaments slender, winged, exceeding the obovate-obtuse anthers, which are 4–5 lines long; capsule elliptical, acutely angled and strongly beaked.

The description is from specimens collected near the type locality, "Pullman, Whitman Co., Washington."

20. Calochortus longebarbatus *Watson*.

PLATE XVI, FIG. 1.

Calochortus longebarbatus WATSON, Proc. Amer. Acad., Vol. XVII, 1882, p. 381.

Near *C. pavonaceus*. Stem slender, a foot or so high, bulbiferous at base, stiffly erect; inflorescence umbellate if more than one flowered, short linear bracts subtending the umbel or a single flower on a stem; true radical leaf one, lance-linear, acute, 2 to 3 lines wide, nearly as long as or exceeding stem, a linear bract-leaf on lower part of stem just above radical leaf; sepals ovate-lanceolate, acuminate, a little shorter than petals, greenish lilac within; petals lavender, lighter below, with a dark purple circular band above gland, obovate-cuneate, denticulate, margin naked; gland small, roundish, covered with densely matted brown hairs, with some long silky hairs above and beside it; filaments slender, winged below, 2 to 3 times as long as anthers; anthers narrowly ovate, obtuse; mature capsule 9 to 10 lines long, strongly winged and beaked.

"In low grassy grounds, Falcon Valley, Klickitat County, Washington Territory."

The description given is of average specimens from the type locality, and from Hood River, Oregon. From the description it will be seen that *C. longebarbatus* closely resembles *C. pavonaceus* except that the bracts are near the base instead of the middle, there are no hairs on the margin of the petals, and the filaments are somewhat longer. The two may run into each other, but to definitely ascertain whether this is the case specimens from a much wider range are needed.

21. Calochortus howellii *Watson*.

Calochortus howellii WATSON, Proc. Amer. Acad., Vol. XXIII, 1888, p. 266.

Stem erect, a foot or more high; radical leaf solitary; cauline leaf narrow and short; sepals ovate and shortly acuminate; petals white, an inch long, denticulate, slightly ciliate near the base, covered within with short crisped hairs, those above the gland denser and dark greenish; gland transversely oblong, densely covered with short yellow hairs; anthers oblong-acute, and apiculate, 3 lines long; capsule elliptical, acute, 9 lines long.

"Found near Waldo, Oregon, in 1884, and at Roseburg in 1887."

The above description is drawn from specimens collected near the type locality.

Group 2. ROCKY MOUNTAIN MARIPOSA.

Gland transverse and narrow, extending across petal from side to side; leaf usually as in Section I.

22. Calochortus gunnisoni *Watson.*

Calochortus gunnisoni WATSON, Bot. King's Rept. Ex. 40th Par., p. 348.

The usually single radical leaf linear-lanceolate, acute, flat, and not channeled; stem bulbiferous at base, a foot or so high, erect; inflorescence usually umbellate, the umbels 1-4-flowered and subtended by two or more rather broadly lanceolate, acuminate bracts; one or several cauline leaves below the umbel; sepals narrowly ovate, acute, with scarious margins, yellowish within, often marked with dark blue; petals creamy white, often banded with dark blue above gland, cuneate, with a broad claw, usually rather truncated above, sometimes slightly rounded, 12-15 lines long; gland narrow, extending transversely nearly from side to side of petal, and covered with short, dense, glandular clavate hairs; anthers equalling filaments, ovate, cuspidately acute; capsule cylindric-triangular.

Described from an average sized specimen from Boulder, Colorado. The species is found on the eastern side of the Rocky Mountains from Nebraska to New Mexico.

"Rocky Mountains of Colorado. Collected also in Utah by Gunnison."

The color of the petals is variable, flowers from different localities showing all the gradations from white, through pink, lilac, purple, and blue, with considerable variety in the color of the bands marking the lower portion of the petal; but a greenish tinge and greenish hairs characterize them all. Some of these color forms have been named.

Group 3. WEED'S MARIPOSA.

Petals covered with slender hairs; capsule narrowly oblong, with thick obtusely angled cells; radical leaf as in *C. albus* (Section 1), solitary, long, shining, and not channeled; bulb heavily coated with coarse black fiber.

23. Calochortus weedii *Wood.*

PLATE XVII.

Calochortus weedii WOOD, Proc. Acad. Nat. Sci., Phila., 1868, p. 169.

Corm deep seated, heavily coated with coarse dark brown fiber; stem 1–3 feet high, stout and flexuous, usually branched, leafy, one to many flowered, not bulbiferous; radical leaf lanceolate-acuminate (the same as in *C. albus*), solitary, broad, shining, flat and not channeled; cauline leaves broad, acuminate; sepals often exceeding petals, narrowly ovate-lanceolate, acuminate, yellowish within, often with a hairy spot at base, with scarious margins; petals cuneate, denticulate, either rounded or more or less truncate above, 12–15 lines long, orange colored and covered with long silky yellow hairs, each set in a dark brown spot, upper quarter usually naked; gland small, circular to oblong, densely matted with short hairs; filaments filiform, exceeding the oblong anthers; capsule narrow, attenuate upward, 1½ inches long.

Described from specimens from San Marcos, San Diego County.

"San Diego."

C. weedii in its various forms is found in the Coast Range of California, from San Luis Obispo County south to Lower California. It is easily distinguished from other Mariposa Tulips by the single large radical leaf (one to two feet long by five to eight lines wide), the very heavy, coarse, stringy, fibrous coating of the bulb, and the long silky hairs springing from the brown dots on the petals.

The species is almost as variable as *C. venustus*, each variation having its own range, where it is found to the exclusion of all others.

Beginning at San Diego, we have the typical *C. weedii*, a large orange-colored flower, covered with yellow hairs and dotted with brown. In this form the petals are usually full, although occasionally somewhat truncated. Nearly all of the *C. weedii* in San Diego County, whether in the interior or on the coast, conform to the type.

23*a*. *C. weedii* var. *purpurascens* WATSON, Proc. Amer. Acad., Vol. XIV, 1879, p. 265. (*C. plummeræ* GREENE, Pittonia, Vol. II, 1890, p. 70). Going north to Los Angeles and San Bernardino, we find a broad belt of country where the flower of *C. weedii* is still full petaled, but the color is lilac or lilac purple. While possessing hardly any

analytical points of difference, it may be said that in cultivation this form proves to be a stronger and hardier plant than the type. A curious fact in connection with the bulb is also worthy of mention. The coarse fibrous coating which the variety has in common with the type is often found to cover not one bulb, as is usual, but two large bulbs of about equal size, laid so flatly face to face that outwardly the bulb retains the round form of the species, and it is only by breaking the coating that the presence of the second bulb is discovered. This habit of the bulb can hardly be called offsetting, as it is impossible to call either the parent bulb. In the San Jacinto Mountains this form varies to pale pink and white. It is the *C. weedii* var. *albus* of horticulture.

A fine plate of this variety may be found in "The Garden," Feb. 2, 1895.

23*b*. *C. weedi* var. *vestus*. Continuing still further north to Santa Barbara, we again find the species, but the petals are much more truncated and curiously fringed with brown hairs, while the color is reddish brown.

23*c*. *C. weedii* var. *obispoënsis*, var. nov. (*C. obispoënsis* LEMMON, Bot. Gaz., Vol. XI, 1886, p. 180.) This is an extreme form found in San Luis Obiśpo County. The petals of the flowers are still more truncated than in the preceding, and near the town of San Luis Obispo, which I think is the northern extreme of its range, the flowers assume a most fantastic form, the brownish petals being so much truncated that the sepals far exceed them, and the hairs which are scattered in typical specimens here seem to be condensed upon the small remaining surface of the petals.

"On dry, stony hills near San Luis Obispo, Cal."

It is, of course, impossible to enumerate here all the many gradations in this plant, which forms one of the most interesting studies in plant variation.

Group 4. GOLDEN BOWL MARIPOSAS.

Petals yellow, lower half covered with clavate hairs; radical leaves long-linear and deeply channeled.

24. Calochortus clavatus *Watson.*

Calochortus clavatus WATSON, Proc. Amer. Acad., Vol. XIV, 1879, p. 265.

Stem stiff, very stout, strongly zigzag, branching, 1–3 feet high, if few flowered umbellate, if many flowered with a terminal umbel and short laterals at the axils of the rather large cauline leaves; the single radical leaf glaucous, linear, deeply channeled, often 1–2 feet long; cauline leaves as broad and prominent; pedicels stout, 2–5 inches long; sepals acuminate, broadly ovate-lanceolate, nearly as long as petals, yellowish within, often greenish without, spotted, with dry scarious margins; petals rather truncate, 18 lines wide, 21 lines long, broadly fan-shaped and strongly arched, with a broad claw, the gland deeply pitted, with yellow hairs; the side of gland and the lower half of petal densely hairy with long yellow hairs, each tipped with a knob-like point, and purplish red at base; color a rich yellow, with claw often reddish brown and a reddish brown band above the hairy zone; perianth bowl-shaped ("formed like a broad-based cup"); filaments slender, a little exceeding the ovate-oblong, obtuse, purplish-brown anthers; capsule narrow, attenuate above and below, 3 inches long.

Described from medium sized plants.

Found on dry rocky points, usually in volcanic soils. The species is widely but peculiarly distributed from Newhall (Los Angeles County) to San Luis Obispo; it is also found in the Sierra Nevada near Pleasant Valley (El Dorado County), and on White Rock, an isolated quartz rock-mass in the plains of Mariposa County. Doubtlessly found also on lava formations at many intermediate points.

"California (San Luis Obispo; J. G. Lemmon, 1878)."

This *Calochortus* is the stoutest stemmed, tallest, and largest flowered of all the Calochorti. The heavy, strongly zigzag stem, yellow bowl-shaped flowers, and clavate hairs, are strongly marked characters shared in by no other member of the genus. The knob-like tips of the hairs are translucent, having the appearance of little icicles. But although the prominent characters remain constant, there is considerable local variation.

In Ventura and Los Angeles counties (Piru City to Newhall), for instance, the flowers are a very rich yellow and the plant is rather low and stout. Near the city of San Luis Obispo, the plant has the same habit, but the upper half of each petal is backed with olive which showing through gives a peculiar changeable color effect. In El

Dorado County the form is very tall and large, often three feet in height, with flowers five inches across, and of a lighter yellow. The hairs are longer and the knob-like tip is smaller.

25. Calochortus concolor, sp. nov.

Calochortus luteus var. *concolor* BAKER, Plate in "The Garden," Dec. 7, 1895.

Bulb large, reddish; radical leaves narrow, glaucous, deeply channeled; one or more cauline leaves below umbel; stem 2 feet in height, one to several flowered, if more than one flowered, umbellate; pedicels stout, 1 to 3 inches long; umbel subtended by linear bracts; all parts of plant very glaucous, lower stem and sepals with a bluish bloom; sepals ovate-acute, with scarious margins, yellowish within, brownish on back; petals a deep rich yellow, tending toward orange, broadly fan-shaped, 15–18 lines long, and as broad as long, slightly rounded above, the lower third densely hairy with long erect yellow hairs; gland small, oblong; anthers yellow, linear, exceeding filaments; capsule strongly triquetrous, lance-linear, attenuate above (imperfect in type specimen).

Described from a large plant collected by Mr. D. Cleveland at Laguna, on the edge of the desert, San Diego County.

This is one of the desert species found in rocky soil in various places on the desert side of San Diego County and in the Cuyumaca Mountains, also at Mill Creek, near San Bernardino, and in Hermit Valley, Riverside County. The writer does not agree with Mr. Baker in referring this species to *C. luteus;* its affinities, if any, are with *C. nuttallii* or *C. clavatus*. Following *C. clavatus* it is the largest flowered, and the showiest of the yellow Mariposa lilies.

Group 5.

Petals nearly naked, gland round, small, and densely hairy with matted hairs; leaves ashy blue, linear, deeply channeled. Desert plants.

26. Calochortus kennedyi *Porter*.

Calochortus kennedyi PORTER, in COULT. Bot. Gaz., Vol. II, 1877, p. 79. Plate in "The Garden," Feb. 11, 1893.

Stem very low, rather stout, often only 1–4 in. high, 2–4-flowered; radical leaves linear, 2–10 in. long, channeled, very glaucous, as are the stem and bracts; flowers produced in an umbel, which is subtended by short bracts; pedicels 3–4½ inches long; sepals ovate-oblong, about equalling petals, obtuse

or acute, with white scarious margins, vermilion inside, often spotted brown near base, brownish without; petals cuneate, rather truncate, 10–15 lines long, not quite as broad as sepals, a dazzling vermilion, naked above, a few scattering hairs below; gland small, round, densely matted with short hairs and bordered nearly black; filaments one-half as long as the oblong-ovate, obtuse, brownish purple anthers, which are 3–4 lines long; capsule 1½–2 in. long, 4–5 lines wide, attenuate above.

Found in the arid regions of southeastern California from near the Ojai Valley (Ventura County) and Tehachapi Station, along the eastern flank of the southern continuation of the Sierras into San Diego County, eastwardly to the Argus Mountains in southwestern Nevada, and throughout Arizona.

"Kern County, California."

In California the flowers are a vermilion color, in Arizona and Nevada, orange. While the stem may reach eighteen inches in height, it often hardly rises above the ground, and the meager ashy foliage gives little promise of the dazzling flower. It is probable that in eastern Arizona and in Nevada *C. kennedyi* will be found merging into *C. aureus*.

27. Calochortus aureus *Watson*.

Calochortus aureus WATSON, Amer. Nat., Vol. VII, 1873, p. 303.

Low, 4–6 inches high, with a single linear carinate radical leaf, 3–4 inches long; scape short, 1–2-flowered, the single pair of bracts linear, 2 inches long; sepals greenish-yellow, with a dark-purple spot near the base, oblong- or ovate-lanceolate; petals broadly cuneate, 15 lines long, bright-yellow, with a small well-defined, circular, densely hairy gland near the base, and a lunate purplish spot above it; young capsule narrowly oblong, not winged.

"On sand-cliffs, Southern Utah."

The writer has no acquaintance with the species except as scant herbarium specimens. The description and locality are the original of Watson.

Group 6. BUTTERFLY TULIPS.

Petals slightly hairy below, usually oculated and brilliantly colored; gland prominent, round or lunate; stem bulbiferous at base, erect, slender, branching; radical leaves, usually a pair, channeled, linear, slightly glaucous, not ashy blue as in the desert species; cauline leaves and bracts narrow.

C. luteus and *C. venustus* are the true Mariposa or Butterfly Tulips. It would be difficult to name any other group of plants in which can be found such a wonderful diversity of color and marking. The colors range from white through lilac to purple, from delicate pink through light reds to deep glowing reds; buffs, yellows and citrons are all present; and the tintings, oculations, and blotchings are bewilderingly numerous.

Watson bases his division of the group on the differences in the gland: that of *C. luteus* being from transversely oblong to narrowly lunate; that of *C. venustus* from round to longitudinally oblong. While the casual observer would place all together, Watson's division is a true one. In *C. luteus* the capsule is acuminate from a triangular base, in *C. venustus*, linear.

28. Calochortus luteus *Dougl.*

PLATE XVI, FIG. 2.

Calochortus luteus DOUGL., Bot. Register, Vol. XIX, 1833, Tab. 1567.

Plants usually dwarfish, 8–10 inches high, stem slender, stiffly erect, bulbiferous at base, often branching; bulblets enclosed within sheath of stem; radical leaves linear, channeled, 1–3 lines wide, bright green, slightly glaucous; cauline leaves or bracts linear, 1–3 inches long, peduncles 1–4 inches long; sepals narrowly ovate-lanceolate, acute, 12 lines long, 2–3 lines wide, yellowish within; petals cuneate, as long as broad, slightly rounding above, yellow or tending toward orange, not oculated, but having penciled lines radiating from gland to center of petal, claw broad; gland rather broad, lunate, densely hairy with ascending matted yellow hairs, a very few scattering hairs above reaching to middle of petal; stamens about equalling style, filaments slender, a little longer than the light yellow, oblong-linear, acute anthers; mature capsule acuminate from a triangular base, 1½–2 inches long.

The description is drawn from strong specimens collected at Monterey.

"California."

The type described of *C. luteus* follows the coast-line from Anderson Valley (Mendocino County) to San Diego County. It grows in adobe soil and is dwarfish. Specimens from Mendocino, San Francisco, Monterey, and San Diego counties are exactly alike.

Farther in the interior, from Sonoma County (Sebastopol) to the Sacramento Valley, and south along the foothills and higher plains of the east side of the San Joaquin Valley, a second form grows, exactly like the type except that it is tall and slender. This is the form commonest in the Sacramento and San Joaquin plains. By slight gradations through a multitude of forms, a dark eye develops, and the habit becomes more luxuriant, until we have *C. luteus* var. *citrinus*.

28*a*. *C. luteus* var. *citrinus*. This variety is merely a color variation of that which follows, the color varying from yellow to deep lemon, and the central spot being darker and not oculated.

This variety is found at its best in the Russian River region from Hopland, Mendocino County, to the town of Sonoma, in Sonoma County. In the latter place, the color is a deep citron and the eye has disappeared. At various other points, for example, eastern Lake County, Linden (San Joaquin County), near Placerville (El Dorado County), and in various localities in the foot-hills of the Sierra this form is approached.

28*b*. *C. luteus* var. *oculatus*. This variety is usually much taller, larger flowered, and often many flowered; petals white to lilac or purple, often 2 inches long, the same in width, the center of each showing an oblong dark brown spot oculated with yellow, and many radiating pencilings; sepals also oculated.

The type of *C. luteus* var. *oculatus* as above described is common in the Coast Range from Shasta County to San Francisco Bay. It is to be expected that it would occur as far south as Monterey County, but the writer has seen no specimens from that region.

In the foot-hills of the Sierras, from Butte to Kern counties, there is a form slightly differing from the type. The petals have less yellow in the middle third, the blotch is smaller, the gland rather deltoid than lunate, and the pencilings are lighter—all trifling differences which only close study will detect; yet they are quite sufficient to separate the *oculatus* of the Coast Range from that of the Sierras.

C. luteus var. *oculatus* of the Sierra foot-hills is very constant. While in some localities it crosses with var. *citrinus*,

specimens from near Chico (Butte County), from Placerville (El Dorado County), and from Dunlap (Fresno County), are identical in every respect. As before stated, *C. luteus* var. *citrinus* and var. *oculatus* hybridize readily, and in many localities where both are found, there are cross breeds in endless variety, running as is usual in such cases to greater extremes than either parent. In Mendocino County, from Ukiah to Hopland, and in El Dorado County, near Camp Creek, these crosses are particularly plentiful and beautiful. There are other strains of *C. luteus*, each found in a more or less extensive territory.

28*c*. *C. luteus* var. *robusta* (*C. venustus* var. *robusta* (Hort.).) In portions of El Dorado County a form approaching *C. oculatus* is found growing in wet grounds (oftener in wire grass lands), which is there dwarfish and with three to five almost spheroid bulblets on the stems. The colorings are very rich; the forbidding surroundings seem to have developed unusual vitality in this form, for in ordinary soil the bulbs produce unusually tall, stout stems but retain the bulbiferous habit.

29. Calochortus vesta *Purdy*.[1]

Calochortus vesta PURDY, The Garden, Oct. 12, 1895, with colored plate; Gardeners' Chronicle, 1895, Part II, page 14.

Tall, large flowered, stem leafy, pedicels elongated, 8–12 inches long; petals more narrowly cuneate than in *C. luteus* var. *oculatus*, white tinged with lilac (with rare albinos), instead of an oculated spot having a broad reddish or dark brown band across the middle; gland narrow, doubly lunate, extending across from side to side of petal; bulblets long and slender, 1 to 4 to the stalk, not enclosed in sheath of stem, but set at an angle.

While *C. vesta* is grouped with *C. luteus* and *C. venustus*, it is a strongly marked form which does not hybridize with either. *C. vesta* is found in adobe soil (sticky black or blue clay) from Sonoma and Napa counties to Humboldt County, California.

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[1] *C. vesta* and the varieties of *C. venustus*—var. *roseus*, var. *purpurascens*, and var. *eldorado*—were first mentioned in a catalogue of bulbs issued by the writer.

C. vesta was first described in an unsigned article in "The Gardeners' Chronicle" (August, 1896). It was so named in honor of the author's wife. In the same article, *C. venustus* var. *roseus* and var. *purpurascens* were also described under the names *C. roseus* and *C. purpurascens*, thus giving specific rank to the catalogue varieties.

A fine colored plate of *C. vesta*, *C. venustus* (the type, known as var. *roseus*), and *C. venustus* var. *purpurascens*, was published in "The Garden" (London); and a good plate of *C. venustus* (type), *C. luteus* (type), and *C. luteus* var. *citrinus*, appeared in the same magazine 1883 (?).

Calochortus bulbs move from year to year by means of a spongy process growing from their base, at the end of which the new bulb is formed. By this means the bulblets of *C. vesta*, being set at an angle, travel some distance before at maturity they take a vertical position. By digging into a patch where *C. vesta* has been undisturbed for some years, the ground will be found literally full of bulbs, the immature ones placed at every angle. In most bulbiferous Mariposas, the bulblets being placed inside the stem sheath, and parallel to the stem, grow straight down and form a close clump.

C. luteus var. *robusta* has a habit somewhat similar to that of *C. vesta*, though not so marked; and in *C. venustus* var. *purpurascens* the habit is exactly the same as in *C. vesta*.

30. Calochortus venustus *Dougl.*

Calochortus venustus DOUGL. ex. BENTH. in Trans. Hort. Soc., Ser. II, Vol. I, 1835, p. 412, Tab. XV, fig. 3.

Stem stiff, erect, usually branching, 4 inches to 3 feet in height, bulbiferous, with a single bulblet at base; one or two linear, radical leaves, 1–3 lines wide, quite glaucous; pedicels 2–5 inches long; sepals oblong-lanceolate, 4 lines wide, 18 lines long, acute, as long as petals; petals broadly obovate-cuneate, slightly rounding above, wider than long, 16 lines long, 20 lines wide, color white to lilac above, carmine below, oculated like *C. luteus*, with reddish pencilings on each side of eye, and a transversely oblong, rose-colored blotch near apex, claw reddish brown; gland roundish or oblong, densely matted; filaments a little exceeding the narrowly oblong, obtuse anthers; capsule linear, 2–3 inches long.

Described from a specimen collected in San Benito County, California.

C. venustus is even more variable than *C. luteus*. While *C. luteus* is almost always found in clay, sometimes heavy clay, *C. venustus* usually grows in light open ground, often in sandy soil.

30*a*. *C. venustus* var. *roseus*, The Garden, Oct. 12, 1895.

The typical form described above is known as var. *roseus*, or *C. roseus*.

30*b*. *C. venustus* var. *eldorado*, var. nov. Differs from var. *roseus* in having the more cuneate petals always narrower than long. Found in all colors from white through lilac to purple, from pink through many shades to deep claret, with or without rose-colored blotch near the top of petal. The plants are stout, usually tall (1–2 feet), branching, and large flowered.

C. pictus (The Garden, Oct. 12, 1895) is merely a white form of the above variety.

30*c*. *C. venustus* var. *purpurascens*[1], The Garden, Oct. 12, 1895.
This is a strong growing variety, like the type but taller; flowers lilac to purple, petals oculated and more leafy, but without the rose-colored blotch. The plants are very bulbiferous, producing 1–4 bulblets a year, which scatter as do those of *C. vesta;* they are not enclosed in a stem sheath.

Found growing in adobe (heavy clay) soils, from San Luis Obispo County to Suisun Bay, California.

30*d*. *C. venustus* var. *sulphureus*, var. nov. As in the preceding, like the type but taller; petals a light, warm yellow, with eye in center, and rose-colored blotch at top.

Occurs at Newhall and Alcalde (Kern County), in the lower end of the San Joaquin Valley.

C. venustus is as widely scattered as *C. luteus*, and has many strains, some local, others widely spread. There are two principal strains. One commences in the Coast Range at Antioch (Contra Costa County) and extends southward into the interior or dryer portion of the Coast Range to Los Angeles County.

The type described, as found from Antioch to Paso Robles, always has a rose-colored blotch at the top of the petal. In this strain the petals are broader than long, giving a very full flower. At Paso Robles there is a break, and we find a wonderful variety of color forms, from white to purple, and from pink to deep red, always with a rose

[1] The original *C. venustus purpurascens* Watson was described from specimens from Kern County, in the southern Sierras, and is without doubt only the purple extreme of the El Dorado strain. The name has for years been used for a purple variety of the Coast Range form, which is distinct in every way. As the latter is perfectly distinct, with features by which it can be readily identified, and the former is a mere color extreme, occurring usually with other colors, the rule of priority could, I think, be shaded a little, and the name be used for the Coast Range type.

blotch at the top of the petal. From this point south to Los Angeles County the forms seem to change frequently.

At Preston and Newsom's Springs (San Luis Obispo County) there is a pretty white form, in color exactly like *C. catalinæ* (white, tinged lilac), destitute of both eye and rose-colored blotch, but with the ovate dark brown spot on the claw of the petal, as in *C. catalinæ*.

At Santa Barbara the type with the rose blotch reappears, and it is found at intervals southward.

In the Ojai Valley and at Newhall there is a white-flowered variety with a rose blotch and narrow petals, closely resembling the white-flowered plants at Paso Robles. It is also tall and slender.

At Newhall a new color strain comes in, quite novel in *C. venustus;* this (var. *sulphureus*) is light yellow, narrow petaled, and has the rose blotch. At Elizabeth Lake, a point about midway between Newhall and Alcalde, the species is again seen in the infinite variety of color found about Paso Robles and in the southern Sierras, but with the additional yellow form.

Newhall is the most southern point from which the writer has specimens of *C. venustus*.

Mr. S. B. Parish has written a very interesting description of these color forms.

The Eldorado strain, so termed by the writer in a catalogue of bulbs, is found in the Sierra Nevada. It begins to appear a little north of Camp Creek (El Dorado County), high up in the foot-hills in the upper Yellow-Pine belt (growing oftenest in open woods), and extends at a nearly uniform altitude in the Sierras south to the Tehachapi and Tejon mountains. The colors of the flowers are richer than those of the Coast Range forms. They vary from white to lilac and deep purple, from pink to deep red, and sometimes light cream flowers are found. The eye is dark brown, not much oculated, the blotch at the top of the petal varies from rose to gold, and flowers can be found with rose or gold rays across the entire top of the petal; but the rose blotch at the apex, instead of being a constant feature as is

usual in the Coast Range forms, is rare. At all points from which specimens of this form have been collected the variety of color is shown to vary greatly in proportion; sometimes nineteen-twentieths of the flowers are white, again red or purple will predominate, and this in localities only a few miles apart; but no single color form is ever found to the exclusion of all others.

Group 7. LILAC MARIPOSAS.

Petals white, lilac, or purplish, not oculated, more or less hairy; gland small, round, and densely hairy; leaves linear, channeled; capsules linear except in *C. catalinæ*.

31. Calochortus splendens *Dougl.*

Calochortus splendens DOUGL. ex BENTH. in Trans. Hort. Soc., Ser. II, Vol. I, 1835, p. 411, Tab. XV, fig. 1.

Stem slender, often bulbiferous at base, usually a foot or two high; sepals often spotted purple, ovate-acuminate, about equalling petals; petals broadly fan-shaped, circular above, 1-5 lines long, 18 lines broad, upper half naked, scattering short hairs around the gland and on lower third, the claw very short; color from lilac to purple, lighter about gland, usually reddish purple on claw; gland small and round with a mound of matted agglutinated hairs which are scarcely distinguishable as hairs, and are frequently absent; anthers purple, obtuse, one-third as long as filaments; capsule linear.

Described from strong plants from San Diego, California. In horticulture the type is known as *C. splendens* var. *atroviolacea*. It is found from San Diego to Santa Barbara counties, and on the coast islands.

31*a*. *C. splendens* var. *montanus*, var. nov. Like the type but very slender, 8–12 inches high, smaller flowered, and more densely hairy about the gland with short yellow hairs; color lilac to salmon pink; often bulbiferous.

Described from specimens from Raynetta, San Jacinto Mountains.

31*b*. *C. splendens* var. *major*, var. nov. This resembles var. *montanus* but is not bulbiferous, is stouter and much larger flowered; petals pale lilac, lighter below, with very long tangled hairs scattered on middle third.

Described from specimens from Monterey County, California. Found also in San Luis Obispo County.

The figure shown in Douglas' plate is that of the form here described as the type. Baker, in his Tulipæ, gives an accurate description of this same form, calling it *Calochortus splendens;* but Watson, in the "Botany of California," has described as the type the form here given as var. *major*. The marked difference between these two forms is in the hairs. In the type the hairs are short and found on the lower third of the petal; in var. *major* they are on the middle third of the petal, and are long and cobwebby. In addition to this, the latter variety is of a stronger habit, has larger, lighter colored flowers, and is without bulblets.

31*c*. *C. splendens* var. *rubra*, a form found in eastern Lake and Napa counties, is singularly enough in gland and hairs almost exactly like the type at the other extremity of the range. It is a very large plant, with a deep-seated yellow bulb, tall (1–3 feet), stout stem, and a large reddish lilac, pink or purplish flower, the petals quite hairy with short hairs on lower third, and with a deep reddish purple claw.

C. splendens var. *montanus* has been mistaken for *C. palmeri*, but it is closely related to the type; in fact, aside from the color of the hairs above the gland, and the more slender habit, it hardly differs from the type except in its habitat. The writer has it from "cienagas" (wet springy spots) near Tehachapi, from wet spots near Bear Valley (San Bernardino County), and from wet meadows in the San Jacinto Mountains. The other three forms always grow in dry, rocky soil.

32. Calochortus palmeri *Watson*.

Calochortus palmeri WATSON, Proc. Amer. Acad., Vol. XIV, 1879, p. 266.

Stem very slender, lax and flexuous, a foot or two high, 1–7-flowered, bulbiferous near the base; sepals with narrowly acuminate recurved tips, spotted; petals 6–12 lines long, white (or yellowish below) with a brownish claw, and with scattered hairs around the ill-defined, broad, densely hairy gland; anthers obtuse, 3 lines long, capsule very narrow, an inch long or more.

"California, near the Mojave River."

The above description and locality are quoted from the original of Watson.

The writer has never seen a specimen which conformed to the description; all of those seen from Southern California collections were either *C. invenustus* or *C. splendens* var. *montanus*.

33. Calochortus catalinæ *Watson.*

Calochortus catalinæ WATSON, Proc. Amer. Acad., Vol. XIV, 1879, p. 268.
Calochortus lyoni WATSON, Proc. Amer. Acad., Vol. XXI, 1886, p. 455.

A foot or two high, general habit as in *C. splendens;* stem bulbiferous at base; sepals broadly ovate-lanceolate, acute, a little shorter than petals, greenish, spotted dark purple on claw; petals cuneate, longer than wide, rounded above, narrowing abruptly to a short narrow claw; color from white tinged with lilac, to lilac purple, with a large ovate purplish-maroon mark, the apex covering claw and the rounded end above gland; gland oblong, densely matted with short agglutinated hairs, a few scattering hairs around and above; filaments slender, about four times as long as the yellowish, oblong-obtuse anthers; capsule oblong, rounding at both ends.

Described from specimens taken near Los Angeles, California.

"Santa Catalina Island." [California.]

C. catalinæ is found from Los Angeles and San Bernardino along the coast to Santa Barbara, and on all the islands along the coast. It is one of the least variable and best marked of all the Mariposas. The oblong capsule is a distinctive feature, while the peculiar ovate mark on the petals is equally characteristic.

The species was first described by Watson from a plant in the capsule, the specimens having been collected on Santa Catalina Island. Later, he described the same species, under the name of *C. lyoni*, from flowers collected at Los Angeles and Newhall by Dr. Asa Gray and W. S. Lyon. *C. lyoni* also grows on Santa Catalina Island, and there is no doubt of its identity with *C. catalinæ*. In an able article in "Erythea," Dr. Davidson of Los Angeles shows that the two species are the same.

34. Calochortus invenustus *Greene.*

Calochortus invenustus GREENE, Pittonia, Vol. II, 1890, p. 71.

Stem very stiff, stout as compared with *C. splendens*, strongly bulbiferous at base, the bulblets large, ovate-oblong as in *C. nuttallii;* radical leaf linear,

deeply channeled; plant very glaucous, as in most desert species; inflorescence umbellate or at least the larger divisions of stem so; umbels, or branch divisions, subtended by three or four lanceolate bracts an inch or two long; sepals ovate-lanceolate, acute, considerably shorter than petals, yellowish green within; petals obovate-cuneate, short clawed, rounding above or abruptly acute, smoky white, claw purplish; gland small, oblong, densely hairy with matted hairs, yellowish short tangled hairs above gland; anthers 2–3 lines long, not equalling filament; capsule 3–4 lines broad in the middle, tapering both ways.

The description given is drawn from specimens taken at Bear Valley (San Bernardino County), California.

"Higher mountains to the westward of the Mojave Desert."

C. invenustus was discovered by the namer, Professor Greene, near Tehachapi. It is also found in the Tejon Mountains, and doubtlessly grows all along the line from there to Bear Valley. Specimens from the former locality are identical with those here described.

The characteristics of the species place it between *C. nuttallii* and *C. splendens*. The prominent offset, stiff, stout stem, and tendency to flower in umbels, are strongly suggestive of *C. nuttallii*, while the flowers resemble those of *C. splendens* var. *montanus*.

35. Calochortus excavatus *Greene*.

Calochortus excavatus GREENE, Pittonia, Vol. II, 1890, p. 71.

Resembling the last [*C. invenustus*], but the bracts ovate-lanceolate, scarious almost to the striate-veined middle portion, their acuminate tips recurved; petals white shaded with lurid purple above, but dark purple below and about the broad obovate hairy gland, which is deeply impressed, appearing like a yellow saccate body on the outside of the petal; stamens as in the last, but anthers dark maroon.

"From Bishop Creek, Inyo County, California, collected by Mr. W. H. Shockley (No. 427)."

Having little knowledge of this species, the writer has quoted the original description and locality of Professor Greene. From specimens seen, the species would seem to come between *C. invenustus* and *C. nuttallii*.

36. Calochortus flexuosus *Watson.*

Calochortus flexuosus WATSON, Amer. Nat., Vol. VII, 1873, p. 303.

Stem slender, very flexuous or almost decumbent, a foot or so high, branching; bracts linear or lanceolate, 6–15 lines long; sepals oblong-lanceolate, greenish with a deep purple spot; petals broadly obovate-cuneate, 12 to 15 lines long, purple, claw deep purple; gland obscure, purplish yellow, scattering glandular hairs above gland; capsule broadly oblong.

The description given is substantially that of Watson.

"Southern Utah and Northern Arizona."

In 1897, the writer received a number of fresh flowers from St. George, Utah, the type locality. They showed the color to be a deep, rich purple (the flowers are sometimes white), the markings varying considerably, showing bands or spots on either petal or sepal; scarcely any two of the flowers were alike in this particular. The very flexuous habit is distinctive. The stems are often even decumbent ("creeping" a correspondent has it). Found also in southern Nevada.

This *Calochortus* grows on the hills in red granite soil. Mr. S. B. Parish collected a species in the desert region of Southern California, which he identified as *C. flexuosus*. This, he writes, grows in tufts of grass in saline meadows. It is, however, hardly probable that the same species grows under such diverse conditions.

37. Calochortus dunnii, sp. nov.

Stem not bulbiferous at base, a foot or two high, slender; leaves linear, deeply channeled; sepals ovate-acute, with white scarious margins, a little over one-half the length of the petals, never recurving, light green without, greenish white within, faintly spotted; petals broadly cuneate, as broad as long, rounded above, white, with a reddish brown transverse band above the gland; gland small and round, densely hairy with short matted hairs, short scattering hairs on each side of the gland only; capsule linear as in *C. venustus*.

Described from specimens flowering in the writer's grounds; they were originally collected near Julian (San Diego County), California, by the veteran naturalist, George W. Dunn, in whose honor the name is given. So

far, this pretty new species is known only from the type locality. It forms a connecting link midway between *C. splendens* and *C. venustus*.

Group 8. GREEN BANDED MARIPOSA.

Petals purplish lilac, with a greenish line down the back, obovate-acuminate.

38. Calochortus macrocarpus *Dougl.*

PLATE XVIII.

Calochortus macrocarpus DOUGL., Trans. Hort. Soc., Vol. VII, 1830, p. 276, Tab. VIII.

Stem bulbiferous at base, stout, erect and rigid, 1-2 feet high, one or more flowered; the single radical leaf, linear, deeply channeled; cauline leaves 3-5, narrow and convolute; sepals about equalling petals, acuminate, purple inside or tinged with purple, darker at base, sometimes spotted and hairy, with a broad scarious margin; petals obovate, narrowly acute or acuminate, or often distinctly cuspidate, a deep purple lilac, lighter at base, with a broad light-colored claw 1½-2½ inches long and half as wide, a greenish line down the middle, the lower third above the gland with scant glandular hairs; gland oblong, densely hairy; anthers purple or yellow, lanceolate, obtuse, 4-6 lines long, about equalling filaments; capsule attenuate upward, 1½-2½ inches long.

In sandy deserts from northeastern California (Modoc and Lassen counties) to eastern Washington and into Idaho.

A very clearly marked species varying but little. There is a white form near Pullman, Washington. The Indians of the northwest are very fond of the bulbs.

Group 9. SEGO LILIES.

Flowers lilac, white, yellow or pink; gland round; stem prominently bulbiferous at base, umbellate.

39. Calochortus nuttallii *Torr. & Gray.*

Calochortus nuttallii TORR. & GRAY., Pacif. R. R. Rept. (Bot.), Vol. II, 1854, p. 124.

Stem erect, stiff, a foot or two high, with a large oblong bulblet at base; inflorescence simply umbellate, 1-5 flowered; radical leaf linear, deeply channeled; cauline leaves 1 to 3, narrow, glaucous, revolute; sepals ovate-lanceolate, with scarious margins, yellowish within, with or without a dark spot at base, which is sometimes hairy, much shorter than petals; petals broadly

obovate-cuneate, with a rather narrow claw, abruptly acute or rounding above, white, yellowish below, with a small round or oblong gland which is covered densely with agglutinated hairs, a few long hairs scattered about the gland; anthers oblong-obtuse, more or less sagittate at base, shorter than filaments; capsule lanceolate, 1–2 inches long.

Described from specimens from southern Utah.

C. nuttallii has the widest range of any *Calochortus*. It is found from the western flank of the Sierras to Nebraska, and from the Snake River to New Mexico. In color it varies greatly and in other characters slightly. The region of its habitat is too little known to enable any one to mark the range of the variations definitely. In Nebraska and eastern Colorado it is yellow; in many places in central Colorado the color varies wonderfully from white and pink to purple; from Salt Lake to Reno, Nevada, and southern Utah, but one form seems to be common, that which is the type here described; at Pine Valley, southern Utah, the flowers are cream colored; and at Dimee, New Mexico, they are orange.

40. Calochortus leichtlinii[1] *Hooker.*

Calochortus leichtlinii HOOKER, Bot. Mag. Tab. 5862.

A dwarfed or alpine form, quite different from any of the other forms, often but an inch or two in height; stem more slender; anthers strongly sagittate; petals smoky white with a dark spot.

Found in the Sierras at an altitude of from 6,000 to 9,000 feet.

There is another variety of this dwarfed alpine *Calochortus* also found in the Sierras, but the description of this form will be reserved until there is fuller material to draw from.

---

[1] *C. leichtlinii* is included under *C. nuttallii* by Watson, in the Botany of California Vol. II, p. 177.

EXPLANATION OF PLATE XV.

Calochortus purdyi.

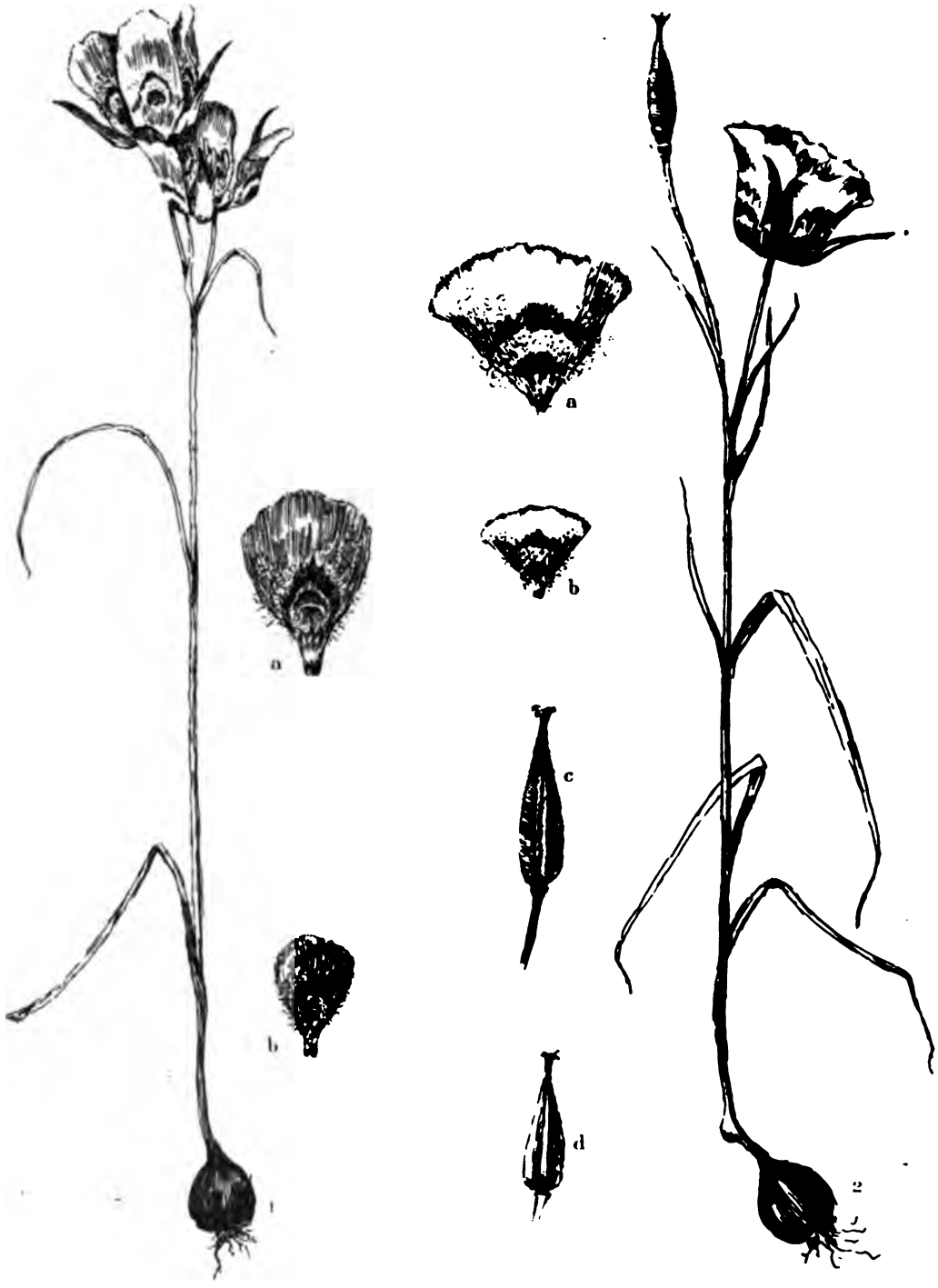
a, Plant, actual size.
b, Petal.
c, Stamen, enlarged.
d, Sepal.
e, Scale on petal, enlarged.
f, Ripe pod.

PHOTO LITH. PAINTON & REY. S.F.

CALLIPHRUM PURDYI

EXPLANATION OF PLATE XVI.

Fig. 1. *Calochortus longebarbatus.*
a, Petal.
b, Petal, less enlarged.
Fig. 2. *Calochortus luteus.*
a, *b*, Petals.
c, *d*, Capsule at different stages of development.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EXPLANATION OF PLATE XVII.

Calochortus weedii.

a, *b*, Capsule at different stages of development.
c, Sepal.
d, *e*, Petals.

PHOTO. 12TH PRINTING & REY. B.

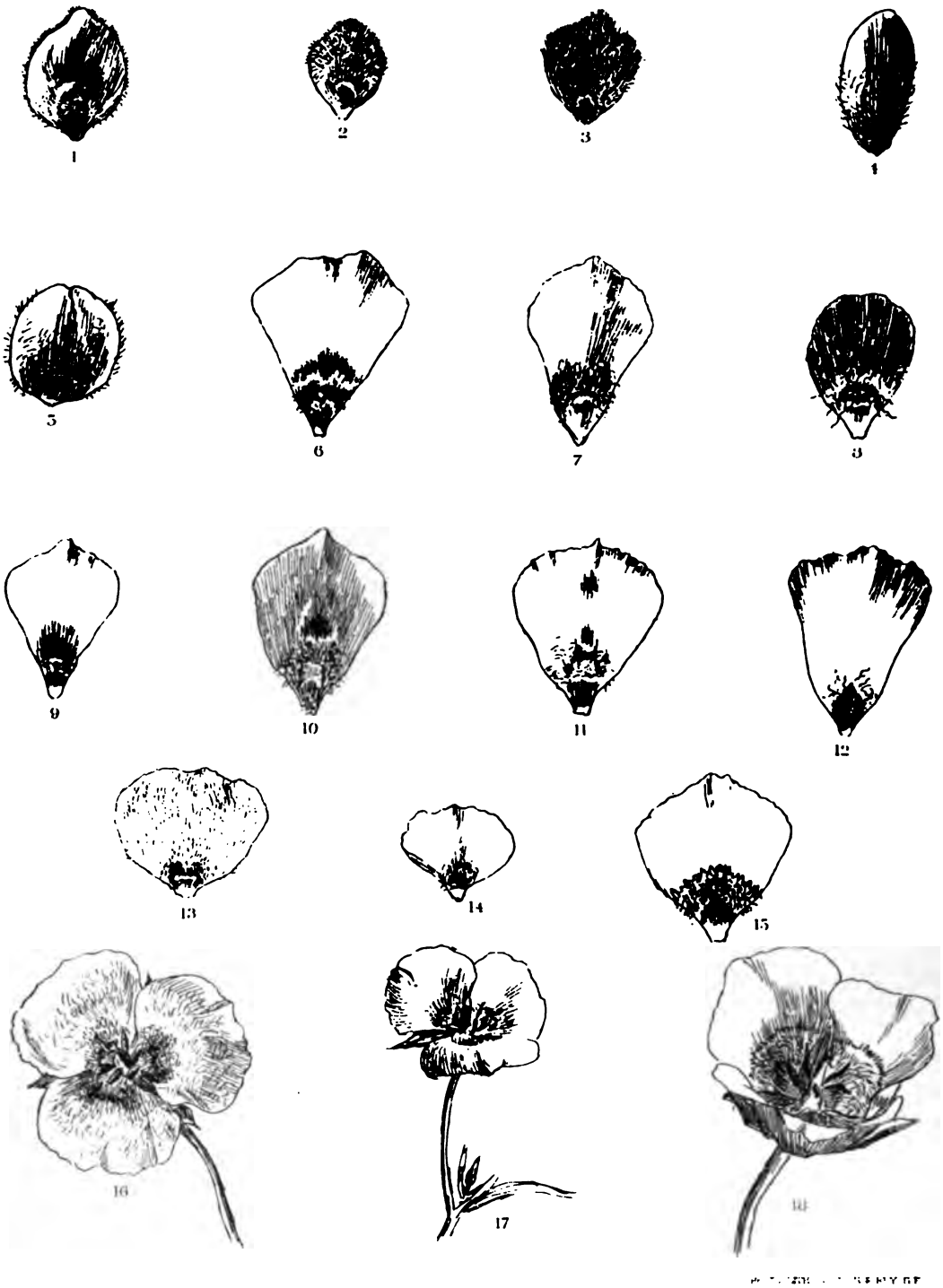
EXPLANATION OF PLATE XVIII.

Calochortus macrocarpus.

EXPLANATION OF PLATE XIX.

Petals of *Calochortus* species.

1. *C. amabilis.*
2. *C. benthami.*
3. *C. maweanus.*
4. *C. amœnus.*
5. *C. albus.*
6. *C. vesta.*
7. *C. gunnisoni.*
8. *C. longebarbatus.*
9. *C. nuttallii.*
10. ***C. venustus.***
11. ***C. venustus roseus.***
12. ***C. catalinæ.***
13. ***C. howellii.***
14. ***C. dunnii.***
15. ***C. clavatus.***
16. ***C. howellii***, flower.
17. ***C. dunnii***, flower.
18. ***C. clavatus***, flower.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A Group of Western American Solanums

BY

S. B. PARISH

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A GROUP OF WESTERN AMERICAN SOLANUMS.

BY S. B. PARISH.

THE first notice in botanical literature of any of the group of *Solanums* which is the subject of the present paper was in 1826, when in the Memoirs of the Academy of St. Petersburg the name *Solanum umbelliferum* was bestowed by Eschscholtz on a plant from "Nova California." As the original description is not easily accessible, a transcription of it, for which I am indebted to Dr. J. N. Rose, is subjoined in the foot-note.[1]

The floral characters here assigned are common to a whole series of plants that the student of Californian botany finds it unsatisfactory to unite, and as difficult to separate on stable lines; nor are the vegetative characters defined with sufficient accuracy to enable one to determine which of these plants the author had in hand.

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[1] 4. Solanum umbelliferum *Esch.*

Solanum umbelliferum ESCH., Mem. Acad. Petersb., Vol. X, 1826, p. 283.

"(Inerme, foliis integerrimis, calycibus quinquedivisis, staminibus æqualibus, inflorescentia terminali.)

"Caule suffruticoso, erecto, foliis ovalibus, acutis, integerrimis, pubescentibus; umbellis terminalibus.

"In fruticetis Novæ Californiæ.

"*Caulis* orgyalis, suffruticosus, fistulosus, angulatus, pubescens; ramis subherbaceis, nutantibus, tomentoso pubescentibus.

"*Folia* alterna, petiolata, ovalia, acuta, integerrima, utrinque pubescentia, vix pollicaria, caulina interdum late ovata sesquipollicaria.

"*Flores* terminales umbellati; umbella plerumque quadriflora, interdum bivel-triflora; involucrum parvum urceolare, integrum, pubescens; pedunculi æquales, elongati, pubescentes. Calyx urceolaris quinquefidus pubescens, laciniis acutis. Corolla calyce triplo major, dilute violacea, quinquefida, extus pubescens. Antheræ flavæ.

"Bacca magna purpurea."

The "*involucrum parvum*" is, of course, the organ better designated by Dr. Gray in the Synoptical Flora as a "cupulate node" at the insertion of the pedicels on the peduncle.

Dunal (1852), in De Candolle's Prodromus, substantially repeated the above diagnosis for the original species, and added to it two others. The significant points in the descriptions of these species are that the one, *S. genistoides*, is characterized as "*ramulosissimis pilosis*," and the other, *S. californicum*, as "*tomentosis candicantibus*." The first of these was founded on a specimen collected in California by Douglas, and the latter on another plant from the same region and collector, and one of Pavon's from "Nova Hispania."

In 1876, Dr. Gray, in a contribution to the Proceedings of the American Academy, reduced Dunal's two species to the original Eschscholtzian *S. umbelliferum*, making the branched hairs of the pubescence the essential character of that species, and at the same time proposing a new species, *S. xanti*, for the reception of certain plants which had since come to hand, and in which the pubescence was of "simple and few-jointed hairs, some of them glandular." No type is specified, but reference is made to specimens collected by Xantus de Vesey, Bigelow, Anderson, and Lemmon. A variety, *S. xanti wallacei*, was proposed for a plant collected on Santa Catalina Island by Wallace.

There are preserved in the Gray Herbarium of Harvard University authentic specimens of all the above species, those representing Eschscholtz's, and Dunal's types coming from the Herbarium of Trinity College, Dublin. By an examination of these it is possible to ascertain exactly what were the plants intended by the different authors, and what more recently collected material may be included with them.

A specimen collected by Hartweg is accompanied with a note in Dr. Gray's hand, certifying it to be an original of Eschscholtz's *S. umbelliferum*. It has a stem moderately hirsute (but not canescent) with a mixture of unbranched and few-branched hairs in about equal proportion. The leaves are ovate, obtuse at base, about two centimeters long, sparsely hirsute, the hairs short and mostly unbranched. All the hairs are unilocular and not glanduliferous.

An original Douglasian specimen represents *S. genistoides*. The stems are slender; the leaves few, somewhat fascicled, minute (5–6 mm.), and ovate; the hairs on the peduncles are mostly branched, on other parts of the plant they are entire, or a few once-branched. As in the first species, all are one-celled and glandless. The specimen has the appearance of coming from a starved plant, as Dr. Gray suggests. Coulter's No. 590, also from Trinity College Herbarium, is a like form.

A specimen of the plant of Douglas, on which *S. californicum* was based, has stout stems canescently tomentose, with hairs all of which are branched. The leaves are broadly ovate (2–4 cm.), cuneate at base, and sparsely hirsute with mingled unbranched and few-branched hairs. The structure of all the hairs is as in the other species. A specimen by Fremont, mounted on the same sheet, is a better representative of this form, and has like characters.

The plant collected by Xantus de Vesey, No. 73, at or near Fort Tejon, California, may be taken as the type of the species which bears his name. It has oblong leaves (circ. 3 cm.) mostly acute at summit and cuneate at base, except a few leaves lobed and truncate at base. The whole plant is moderately hirsute with rather short plurilocular hairs. Anderson's, Lemmon's, and Bigelow's plants are on the same sheet. The first is quite like that of Xantus; the other two are mere fragments with similar pubescence and oblong leaves, which are obtuse or subcordate at base, and probably were taken from plants whose lower leaves were cordate. Wallace's Santa Catalina Island specimen (type of the variety *wallacei*) is likewise fragmentary. It consists of the summit of a branch with an ovate leaf (9 cm.) and a few young leaves, and an umbel of flowers with large corollas (4 cm.); the pubescence of the stem is of long tawny hairs, multilocular and viscid-glandular; the leaf is nearly glabrous above and tawny-villous below. All the hairs appear to be unbranched, but other specimens commonly show some few-branched hairs.

From the above notes on these type specimens it will be seen that the character which has been relied upon as distinctive of *S. umbelliferum*, namely, "hairs branched," is only partially applicable, not one of them being without a considerable admixture of unbranched hairs. Indeed, I have not been able to find a single specimen of this species, among the many examined, in which more or less unbranched hairs could not be detected. On the other hand, many plants are found with a few, often a very few, branched hairs intermixed with the prevalent simple hairs. Moreover, the character of the pubescence, at least in most specimens of *S. umbelliferum*, is different on the various parts of the same plant. On the young stems, notably towards the tips, it is exclusively, or nearly so, of many-branched hairs, while on the leaves, notably on the older ones, it is largely, sometimes exclusively, of unbranched hairs.[1]

A more satisfactory character is found in the structure of the hairs. In *S. umbelliferum* these, whether branched or unbranched, are without cell divisions and are not glanduliferous. In the plants which have been referred to *S. xanti* the hairs consist of elongated cells, some of the cells usually evacuate and collapsed, or atrophied, so that the hairs have a peculiar unevenness; many of them are tipped with black globular glands, causing the plant to be more or less viscid. In glabrate forms the hairs are very short and mostly reduced to a single cell, but they remain glanduliferous. Unfortunately, this character is not entirely constant, and it is possible to find specimens on which there are branched hairs which are also plurilocular and glanduliferous.

The leaves of the group exhibit a wide range of variation in shape, passing from orbicular to oval, oblong, elliptical, and even lanceolate; the apices are either acute or rounded,

[1] In a specimen collected by Kellogg and Harford near Bear Harbor, on the northern coast of California, a third form of hair occurs on the oldest stems. These are densely tomentose with the usual mixture of branched and unbranched hairs, from which stand out scattered spinose branched hairs, 2–4 mm. long.

the bases attenuate, cuneate, rounded, truncate, or cordate. Often a part of this range of variability may be seen on a single plant, and series of specimens can be arranged readily passing by intergradations from one extreme to the other. A tendency to segmentation is manifested by the occasional occurrence on a specimen of a few leaves with a pair, or even two pairs, of basal lobes. The leaves are either smooth or papillose-roughened beneath the pubescence, and are either entire or crenate margined, the latter character appearing to be constant and of some diagnostic value.

The floral characters are practically the same throughout the group, except that *S. wallacei* usually has a narrower and deeper lobed calyx than the others. The corollas are from two to five centimeters in diameter, and in color vary from light to very dark violet, the centers having green markings; the long, bright yellow anthers are sagittate at base, and are on very short, stout filaments; the style exceeds them one-half to one-third its length, is usually straight, but the included portion is sometimes bent. The fruit is very little known; it may possibly afford some satisfactory characters when better understood. It is a smooth, globular, many-seeded berry, about two centimeters in diameter, or perhaps sometimes smaller. In *S. wallacei* it is certainly dark purple when ripe, and this color has usually been assigned to the fruit of all the members of the group; but according to Professor Greene[1] the ripe fruit of *S. umbelliferum* is "yellow;" that of *S. xanti* I have never seen more than a light or whitish green when apparently ripe.

Such being the generally inconstant characters of this group of plants, it is evident that their satisfactory segregation is a matter of no little difficulty. The exercise of that botanical industry which multiplies "species" by the minute description of individuals might reap here an abundant harvest. On the other hand, a rigorous insistence on sharp

[1] Man. Bay Region, p. 268.

and absolute delimitations, which should exclude all intergrading forms, would reduce the whole group to a single heterogeneous species, a disposition, it is safe to say, unacceptable to any one who has studied these plants. Avoiding these two extremes, I have attempted a classification with reference to certain diverging lines of development which are manifested, without insisting upon definite cleavages, which do not exist. Such differences, as every botanist is aware, are more easily perceived in the examination of copious material than characterized. In the present instance the facts as they are in nature are represented by such a treatment; for we have here a series of plants in which variation has outrun the processes of selection, and in which the connecting forms yet remain to unite the diverging lines of evolution. It has also the advantage of preserving the species recognized by Dr. Gray, who with his accustomed discernment fixed upon the best diagnostic character, that of the pubescence, although it does not possess the definiteness he seems to have supposed.

To the group I have prefixed an Arizona plant with a very different corolla, but with further characters shared by the other members of it.

KEY TO THE GROUP.

Corolla deeply 5-cleft; nodes of the peduncle obsolescent. 1. *S. arizonicum.*
Corolla angulately 5-lobed; nodes cupulate, prominent.
Leaves mostly linear-lobed at base. 2. *S. tenuilobatum.*
Leaves seldom lobed.
Plants viscidulous; hairs unbranched.
Leaves crenate; corollas large.
Stems long-hirsute. 3. *S. wallacei.*
Hispidulous or glabrescent. 4. *S. wallacei viridis.*
Leaves with entire margins.
Acute or merely obtuse at base. 5. *S. xanti.*
Cordate or subcordate at base. 6. *S. xanti intermedium.*
Attenuate at base, small. 7. *S. xanti glabrescens.*
Plants not viscid; hairs branched.
Stems villous. 8. *S. umbelliferum.*
Stems canescently tomentose. 9. *S. umbelliferum californicum.*

" *Suffrutescent or suffruticose plants; peduncles lateral, or by the suppression of the growing apex apparently terminal; styles clavate; fruit a many-seeded berry.*

← *Corollas small, 5-cleft; peduncles slightly thickened at the articulation of the pedicels.*

1. Solanum arizonicum.

Barely suffruticose or even herbaceous; stems 3 m. high, not striate or angled, pubescent with unbranched hairs, the upper part canescent, as are the lower surfaces of the leaves; leaves ovate-lanceolate, 2–3 cm. long, prominently anastomose veined, the lower half of the margins coarsely toothed; flowers in small corymbs (about 7-flowered); peduncles surpassing the leaves; pedicels short, 2–5 mm.; calyx 3 mm. high, the lobes ovate; corolla light purple, pubescent without, 5–6 mm. wide, 5-cleft nearly to the base into ovate-acuminate lobes; anthers 3 mm. long, on filaments 1 mm. long; style hirsute below; fruit not seen.

Habitat: Hot Springs, Arizona (397 Toumey, June 17, 1892 [N]).[1]

← ← *Corollas rotate, angulately 5-lobed, violet, with green markings at base; peduncles thickened into a cupulate node at the articulations of the slender pedicels.*

↔ *Pubescence of several-celled, unbranched hairs.*

2. Solanum tenuilobatum.

Suffrutescent, stems slender, angled, glabrescent below, hirsutulous above with short, one- to several-celled, non-glanduliferous hairs; leaves linear to narrowly oblong, 2–3 cm. long, the midrib prominent, all but the uppermost with a pair of hastate linear lobes at base; umbels 1–4-flowered; corolla 12–15 mm. wide; fruit not seen.

Habitat: Mexico,—Lower California (probably near Ensenada, C. C. Parry, April, 1882, type [G]); (Carrizo Creek, Brandegee, April 19, 1893 [A]).

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[1] The letters in brackets denote the herbaria in which specimens are deposited. [A] Herbarium California Academy of Sciences, San Francisco; [G] Gray Herbarium, Harvard University; [N] National Herbarium, Washington; [P] Herbarium of S. B. Parish, San Bernardino, Calif.; [U] Herbarium University of California.

It is a pleasure to record my thanks to Miss Alice Eastwood, Dr. B. L. Robinson, Dr. J. N. Rose, and Dr. Willis L. Jepson, for the opportunity of examining the collections of which they are the custodians. I am also under obligations to Mr. H. M. Hall for specimens and other favors.

3. Solanum wallacei.

Solanum xanti wallacei GRAY, Proc. Am. Acad., Vol. XI, 1876, p. 90; in BREW. & WATS. Bot. Calif., Vol. I, 1876, p. 539; Syn. Fl., Vol. II, Pt. 1, 1878, p. 229. GREENE, Bull. Cal. Acad. Sci., Vol. I, 1885, p. 226; id., Vol. II, 1887, p. 408. LYON, Bot. Gaz., Vol. XI, 1886, pp. 204, 334, 336. BRANDEGEE, Zoe, Vol. I, 1890, p. 143. FRANCESCHI, Zoe, Vol. IV, 1893, p. 137. DAVIDSON, Pl. Los Angeles Co., p. 21 (1896). TRASK, Erythea, Vol. VII, 1899, p. 140.

Solanum xanti WATSON (not GRAY), Proc. Am. Acad., Vol. XI, 1876, p. 117.

Suffrutescent, "often forming round masses;" stems about a meter long, densely tawny-villous with long, multilocular, viscidly glanduliferous hairs which are unbranched, or usually a few once-branched; leaves thickish, sometimes pustulose, usually less densely villous than the stems, crenate margined, the lower ample, cordate, the upper ovate, rounded, or subcordate at base; calyx narrowly funnel-form, deeply cleft, or wider and less deeply divided; corolla 2–4 cm. wide; style glabrate, or villous below; ripe fruit dark purple.

Habitat: Islands off the Coast of California and Lower California, and near the seacoast in Central California.

California (Brandegee [G]); Central California (429 Palmer [A]);

Los Angeles County (Santa Catalina Island, Wallace, type [G]; 76 Lyon, 1885 [G]; McClatchie, Nov., 1893 [P]; Mrs. Trask, Dec., 1894 [A, N]);

Santa Barbara County (Santa Cruz Island, Greene, 1886 [A]; U. S. S. Albatross, Feb., 1889 [A]) (Santa Rosa Island, Brandegee [A]) (Santa Barbara [no collector's name] Jan., 1892 [A]) (Santa Inez Mountains, Brandegee, 1888 [A]);

San Louis Obispo County (Mrs. Blochman, May, 1893 [A]; 428 Palmer, June, 1876 [A]);

Marin County (Miss Eastwood, Oct., 1896 [A]).

Mexico (Guadaloupe Island, 62 Palmer, 1875 [G]; Greene, April, 1885 [P]; Franceschi, Jan., 1893 [A (much reduced form), N, P, U]).

4. Solanum wallacei viridis.

Stout, erect, glabrate, or above hispidulous; hairs mostly reduced to a single cell; leaves ovate, cordate, or rounded at base; calyx cup-shaped, with short lobes.

Habitat: Central California, near the coast.

California (586 Coulter [G]);

Monterey County (Pacific Valley, Miss Eastwood, May, 1897, type [A, P]) (Santa Lucia Mountains, Willow Creek, R. A. Plaskett, Feb., 1898 [A]);

Marin County (Mt. Tamalpais, Miss Eastwood, May 30, 1896 [A]).

5. Solanum xanti *Gray.*

Solanum xanti GRAY, Proc. Am. Acad., Vol. XI, 1876, p. 90; in BREW. & WATS. Bot. Calif., Vol. I, 1876, p. 539; Syn. Fl., Vol. II, Pt. 1, 1878, p. 229. DAVIDSON, Pl. Los Angeles Co., p. 21 (1896). MCCLATCHIE, Fl. Pasadena, p. 641 (1895). COVILLE, Death Valley Rept., pp. 167, 251 (1893).

Stems slender, 3-10 dm. long, woody, or at high altitudes herbaceous from a lignescent base, the younger angled, moderately villous, with many-celled, unbranched hairs, most of them gland-tipped; leaves ovate, ovate-oblong to oblong-lanceolate, 1-4 cm. long, acute or obtuse at base; corolla 1-2 cm. wide; mature fruit apparently light green in color.

Hairs with some of the cells atrophied, and sometimes a few are once-branched.

Habitat: Throughout California, except in the desert region, ascending to 6,500 feet altitude in the mountains, and reaching the borders of Arizona and Lower California.

California (428 Palmer, 1876 [N]; 186 Thomas Bridger [N], a canescently tomentose form); central California (429 Palmer, 1876 [N]) Sierra Nevada Mountains (Lemmon, 1875 [N]);

Alameda County (Piedmont, F. W. Koch, March, 1895 [U]);

Calaveras County (1358 Davy, May, 1898 [P, U]. "Viscid to the touch, herbage malodorous;" leaves thin, neurose);

Fresno County (Fort Miller, Heermann, July, 1853 [N]) (Toll House, 2,050 feet altitude, 7 Hall & Chandler, June, 1900 [P]) (Pine Ridge, 5,000 feet altitude, 93 Hall & Chandler, June, 1900 [P]) (Dinkey Creek, 5,300 feet altitude, 355 Hall & Chandler, June, 1900 [P]) (North Fork King's River, 6,000 feet altitude, 448 Hall & Chandler, July, 1900 [P]);

Inyo County (Willow Creek, 789 Coville & Funston [G, N, P]);

Kern County (Fort Tejon, 73 Xantus, 1857-8 [G, N]);

Los Angeles County (San Gabriel, 108 Brewer [G], the specimen in [U] under this number is of the var. *glabrescens*) (Borders of Mojave Desert, Antelope Valley, Pringle, May, 1882 [N]) (Elizabeth Lake, 1888 Parish, June, 1887 [P]) (Saugus, Brandegee [A]) (Antelope Valley, 2343 Davy [U]);

Mariposa County (Yosemite Valley, Mrs. Dodd [U]);

Mendocino County (Eel River, W. G. Wright, 1894 [A]);

Placer County (Truckee River, July, 1886, 49 Sonne [A]; 398 Sonne [P]);

Plumas County (Mrs. Austin [G]);

San Bernardino County (Bear Valley, altitude 6,500 feet, H. M. Hall, July, 1899, [P]; 3382 Parish, June, 1894 [N]) (Bloomington, Parish, March, 1897 [P]) (Reche Cañon, Parish, 1897 [P]);

Santa Barbara County (Bartlett Cañon, 131 Rothrock, 1875 [G]);

Santa Clara County (Saratoga, 254 Davy, Sept., 1893 [U]);

Sierra County (Webber Lake, Lemmon [G]);

Sonoma County (Freestone, Miss Eastwood, March, 1899 [A]);

Tulare County (Mineral King, Brandegee [A]).

Arizona (Palmer, 1869 [N]) (Central Arizona, 427 Palmer, 1876 [N]) (Fort Apache, 607 Palmer, June, 1890 [G, N]).

Mexico,—Lower California (Ensenada, 3711 Jones, April, 1882 [G], a transition to var. *glabrescens*) (San Pedro Martir, Brandegee, May, 1893 [A]).

6. Solanum xanti intermedium.

Solanum xanti COVILLE (not GRAY), Death Valley Rept., p. 257 (1893).

Stems woody, lax, up to 2 m. long, viscid, leaves cordate to oblong, at least obtuse at base, 3–15 cm. long; corollas 2–4 cm. wide.

Few-branched hairs are often present, indicating a transition to *S. umbelliferum*; while in size and shape of leaf this form passes into *S. wallacei* through its variety *viridis*.

Habitat: California, from Sonoma County southward, chiefly in the foot-hills, but ascending the mountains to 8,000 feet altitude in Southern California.

California (Bigelow [N]; Chas. Sayre, 1875 [N]);

Kern County (Havilah, 1064 Coville & Funston, June, 1891 [N]);

Los Angeles County (Cucamonga, Bigelow, 1853–4 [G]) ("Cahuenga Pass," 189 Brewer [G]) ("San Fernando Valley," 189 Brewer [U]) ("San Fernando Plains," 207 Brewer [U]) ("Santa Susana Mountains," 207 Brewer [G]) (Compton, McClatchie, 1897 [P]) (Pasadena, Mrs. Brandegee [A]);

Marin County (Redwood Cañon, Miss Eastwood, March, 1896 [U]);

Monterey County (Santa Lucia Mountains, 440 G. R. Vasey, 1880 [N]; 20 R. A. Plaskett, Feb., 1898 [N]);

Napa County (Jepson, May, 1897 [P, U]);

Riverside County (Santa Ana River, 141 H. M. Hall, May, 1895 [P]);

San Bernardino County (San Bernardino, 441 G. R. Vasey, 1880 [N]); 4388 Parish, May, 1897, type [G, N, P, U]) (San Antonio Mountains,—Lytle Creek, altitude 5,750 feet, H. M. Hall, June, 1899 [N, P]; Swarthout Cañon, altitude 6,500 feet, H. M. Hall, June, 1899 [G, N, P, U]);

Santa Barbara County (Santa Inez Mountains, G. W. Dunn, May, 1891 [A];

Sonoma County (191 Samuels [N], a form with shorter hairs);

Tulare County (Long Meadow, altitude 8,000–9,000 feet, 206 Palmer, June, 1888 [N]).

7. Solanum xanti glabrescens.

Stems woody, slender, 10–15 dm. long, glabrate, or above hirsutulous with short, mostly one-celled hairs; leaves smaller (2–6 cm.), oblong, elliptical or lanceolate, mostly attenuate or acute at base; corolla 2 cm. wide.

Habitat: From southern Oregon, throughout California (excepting the desert region), to northern Arizona and Lower California. Also doubtfully reported from New

Mexico. This is the most widely distributed form, but apparently confined to lower altitudes. Stems usually lax, and seeking support from other shrubs, but in open ground, notably near the coast, forming low, compact clumps.

Oregon (Josephine County, Howell, May, 1884 [G]).

California (294 Fremont, 1846 [G]; "R. N. A." 1896 [N]) (Sierra Nevada Mountains, Lemmon, 1875 [N]) Southern California (285 Parry & Lemmon [G]);

Alameda County (Berkeley, April, 1900, H. M. Hall [P]);

Butte County (Clear Creek, 191 H. E. Brown, April, 1897 [A, N]) (Little Chico, Mrs. C. C. Brown, April, 1897 [A]);

Calaveras County (Mokelumne Hill, 83 Blaisdell [A]);

Colusa County (Epperson's, Mrs. Brandegee [A]);

Lake County (Mrs. Brandegee, July, 1884 [A]) (Snow Mountain, Mrs. Brandegee [A]);

Los Angeles County (San Gabriel Cañon, 108 Brewer [U]) (Compton, McClatchie, 1896 [P]);

Modoc County (Goose Valley, M. J. Baker [U]) (Little Hot Springs Valley, Baker & Nutting, July, 1894 [U]);

Monterey County (Santa Cruz, 2223 M. E. Jones, June, 1881, in part [A]);

Napa County (Zem Zem, Jepson, July, 1892 [U]) (Vaca Mountains, R. H. Platt, March, 1898 [A]);

Placer County (Mrs. M. M. Hardy, 1893 [A]) (Applegate, Mrs. H. Smith [A]);

Riverside County (San Jacinto River, 3115 Leiberg, March, 1898 [N]);

San Bernardino County (San Bernardino, 4384 Parish, May, 1897, type [G, N, P, U]);

San Diego County (San Diego, Cleveland, 1874, 1875 [G]; April, 1881 [P]; Dec., 1883 [A]; Mrs. Brandegee [A]; Greene, March, 1885 [A], a very leafy form; Miss Cummings, April, 1896 [G]) (Alpine, Mrs. Brandegee [A]) (Temecula Cañon, Greene, 1885 [A]) (Fallbrook, Parish, Nov., 1891 [P]) (Witch Creek, Alderson, May, 1894 [P]) (San Isabel, A. W. Henshaw, April, 1893 [N]; H. M. Hall, May, 1899 [P]) (Oceanside, 4437 Parish, June, 1897 [A, G, N, P, U], a compact, maritime form) ("Southwestern

part of Colorado Desert," Orcutt, April, 1889 [N], but an error in locality is probable);

Siskiyou County (Yreka, 877 Greene, June, 1876 [G]).

Arizona (Fort Mojave, Cooper [G]).

New Mexico ("Chiefly in the Valley of the Rio Grande, below Doña Ana," 1011 Mexican Boundary Survey [N], perhaps an error of locality).

Mexico (Lower California, Pringle, April 6, 1882 [N]. A transition to *S. tenuilobatum*).

↔ ↔ *Pubescence of one-celled hairs, at least those of the stems mostly many-branched, not gland-tipped.*

8. Solanum umbelliferum *Esch.*

Solanum umbelliferum ESCH., Mem. Acad. Petersb., Vol. X, 1826, p. 283. DUNAL in DC. Prodr., Tome XIII, 1852, p. 86. GRAY, Proc. Am. Acad., Vol. XI, 1876, p. 90; in BREW. & WATS. Bot. Calif., Vol. I, 1876, p. 539; Syn. Fl., Vol. II, Pt. 1, 1878, p. 229.

Solanum genistoides DUNAL, in DC. Prodr., Tome XIII, 1852, p. 86.

Suffrutescent or suffruticose, stems slender and erect, 1 m. or more in length, moderately hirsute, the hairs glandless and without cell divisions, mostly branched, but some simple, and these often predominating on the leaves; leaves thin, ovate to oblong, obtuse or somewhat acute at base.

Habitat: Coast counties of central California as far south as Santa Barbara. A doubtful form from Mexico.

California (Hartweg, type [G]; 587 Coulter [G]; Bloomer [G]; Kellogg [G]; Douglas, type of *S. genistoides* [G]; 590 Coulter, same form [G]);

Alameda County (Berkeley, Greene [A]; McLean [U]) (Sunol, Congdon, May, 1892 [P]; Jepson, March 9, 1900 [P]);

Mendocino County (Bear Harbor, 717 Kellogg & Harford, July, 1869 [N]);

Monterey County (Fremont, Jan. 31, 1846 [G]);

San Francisco County (Bigelow, 1853–4 [G]; Kellogg [G]; Bolander, 1866 [N]);

San Mateo County (Bolander, 1892 [G]) (Crystal Springs, Miss Eastwood, April, 1896 [A, U]);

Santa Clara County (Stanford University, 104 C. Rutter, Feb. 10, 1892 [N]);

Santa Cruz County (Santa Cruz, 2223 M. E. Jones, in part [A]) (Glenbrook, Santa Cruz Mountains, H. Davis, April, 1899 [A]).

Mexico (San Martin Island, 30 Anthony [G, N]. Nearly leafless, leaves small, orbicular, trifoliately lobed; probably distinct).

9. Solanum umbelliferum californicum.

Solanum californicum DUNAL, in DC. Prodr., Tome XIII, 1852, p. 86.
Solanum umbelliferum GREENE (not ESCH.), Man. Bay Reg., p. 267 (1894).

Stems stout, erect, densely and canescently tomentose with many-branched hairs, those of the leaves sometimes in part unbranched; fruit said to be "yellow" when mature.

Habitat: Coast Mountains from San Francisco to Santa Barbara, and possibly on the borders of Nevada.

California (Douglas, type [G]; Fremont, 3d Exped. [G]; 589 Coulter [G]; Brandegee [G]) (Santa Maria Mountains, Mrs. Watts [A]);

Contra Costa County (Oakly, March, 1900 [P]);

Fresno County (Alcalde, Mrs. Brandegee [A]) (Huron, Miss Eastwood, May, 1893 [A]);

Monterey County (Monterey, Fremont, 3d Exped., Jan. 31, 1846 [N]; 633 Brewer [G]; 633 Guirardo [U]; Brandegee [A]) (Pacific Grove, Tidestrom, Jan., 1893 [U], a stout form with ovate leaves, 3–4 cm. long, same as Guirardo's plant) (Nacimiento River, Miss Eastwood, May, 1897 [A]) (Los Burros Trail, Miss Eastwood, May, 1897 [A]);

San Benito County (New Idria, Miss Eastwood, May, 1893 [A]);

San Francisco County (Lone Mountain, 14 Kellogg & Harford [A]);

Santa Barbara County (San Rafael Mountains, H. C. Ford, 1887 [G]) (Howard Cañon, Miss Eastwood, May, 1896 [A], albino) (Duford's Ranch, Miss Eastwood, May, 1896 [A]) (Sespe, May, 1897, F. W. Hubby [P]).

Nevada (Carson City, Anderson, 1865 [G], but an error in the locality label is probable).

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An Account of the Species of Porphyra Found on the Pacific Coast of North America

BY

HENRI T. A. HUS

WITH THREE PLATES

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BY HENRI T. A. HUS.*

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I. HISTORY.

FROM a systematic point the genus *Porphyra* has been scantily dealt with. Created in 1824, by C. A. Agardh, to contain those species of "*Ulva*" which possess a red coloring matter, it has since been fully treated by but two authors—J. G. Agardh (1882) and J. B. de Toni (1897). The former author was the first to distinguish between monostromatic and distromatic species. This idea was carried still farther by Kjellman, who in 1883, in his "Algæ of the Arctic Sea," distinguished between the genus *Porphyra* and the subgenus *Diploderma*, the latter to include all distromatic species.

The name *Diploderma* was changed by de Toni (1897) to *Wildemania*.

L. Kolderup Rosenvinge (1893) used the name *Porphyra* for both monostromatic and distromatic species, retaining *Diploderma* as a subgeneric name. This appears

*Contributions from the Botanical Laboratories of the University of California and presented in partial fulfillment of the requirements for the degree of M. S., May, 1899. Prepared under the direction of Professor W. A. Setchell.

to be a step in the right direction, since, as Rosenvinge points out, the distromatic species are frequently monostromatic in portions of the frond. The writer has often found specimens of *P. minuta*, *P. leucosticta* and *P. atropicola* which were partly monostromatic, partly distromatic in the purely vegetative portions of the fronds.

Even though the distromatic character is far more constant than was originally supposed, it seems to the writer there exists no sufficient reason to subdivide the genus, since the plants agree so entirely in habit and external characters as to be readily recognized by the collector as belonging to the genus *Porphyra*.

Other contributors to our knowledge of the genus *Porphyra* are Foslie (1890) and Steinbelt (1886), both of whom chiefly investigated the waters of Northern Europe.

The species of the genus *Porphyra* in Asiatic waters have been little studied. Suringar (1870) mentions *P. vulgaris* as occurring in Japan. Afterwards Kjellman (1897) studied the species of *Porphyra* of the coast of Japan, and in his paper enumerates six new Japanese species.

One of the first to mention *Porphyra* in America was Ruprecht (1852). In an account of a species of *Phyllospadix* collected by Wosnessenski near the mouth of the stream Slavjanka (Russian River), he refers to a parasitic species of *Porphyra*, occurring on the blades: "gegen die Blattenden zu, finden sich kleine parasitirende Exemplaren von *Porphyra*." From the fact that *P. naiadum* And. is the only species of *Porphyra* occurring with any regularity on *Phyllospadix*, it is more than probable that this is the species referred to.

Harvey (1858) in his account of American Algæ, mentions but a single species, *P. vulgaris*, found on both the east and west coast. He is inclined to unite *P. vulgaris* Ag., *P. laciniata* Ag., *P. purpurea* Ag., *P. linearis* Grev., and *P. amethystea* Kütz. under the name *P. vulgaris*. Later, Farlow (1881) described *P. laciniata* Ag. as a cosmopolitan species, and mentions *P. leucosticta* Thur. as probably occurring in New England, but not yet certainly observed.

Collins (1882) reported *P. laciniata* from the east coast, and later (1884) *P. leucosticta* and *P. miniata*.

J. Agardh (1882) reports *P. coccinea (P. naiadum?)* and *P. perforata* from the Pacific Coast. Since then Dr. Anderson (1892) has added two new species to the number, *P. naiadum* and *P. nereocystis*.

Miss Tilden in Century III of "Algæ of North America" has distributed four species of *Porphyra* from the Pacific Coast under the names *P. miniata*, *P. naiadum*, *P. leucosticta*, and *P. laciniata*.

Up to 1898, however, there had been reported from the west coast of North America but four distinct species.

In the winter of 1897 Professor Setchell suggested that the writer investigate the peculiar base of *P. naiadum* And., but specimens of *Porphyra* gathered on collecting trips, and a consideration of those in the herbarium, showed such a variety of morphological and anatomical characters that the desirability of a collection and investigation of the species of *Porphyra* occurring on this coast became apparent; it is the results of these investigations which are set forth in the following paper.

II. MORPHOLOGY.

The shape of the fronds of the various species of *Porphyra* is exceedingly variable, but that of most of them can be reduced to the elongated type of frond. The variation is between linear and oblanceolate, and nearly all the species mentioned in this paper exhibit both characters at various periods of their existence. A striking exception to this rule is *Porphyra perforata* f. *lanceolata*, which, as a rule, is constantly linear; yet there are specimens in our herbarium which are decidedly lanceolate. On the other hand, I believe there are but few mature specimens of *P. tenuissima*, if any, which ever exhibit a linear form. The nearest approach the plant makes to the linear form is when young; it then possesses an oblong outline.

Closely connected with the shape of the fronds are their length and width. These three characters seem to be

closely related and are evidently determined by the same conditions, viz., age, zone, and locality. It would be difficult to say which of these three agencies exerts the greatest influence on the plants. All seem to be of equal importance. A very good illustration is yielded by *P. perforata*. While young specimens of *P. perforata* (three to four centimeters long) are usually irregularly expanded, with a tendency towards the orbicular, we notice that very soon a change in the shape of the frond takes place. As a rule, specimens five centimeters or more in length already show the type, lanceolate with undulate margin; but older specimens, and this applies particularly to those found in the lower part of the litoral zone, and in the upper part of the sublitoral zone, possess a great width and are frequently much lobed and laciniate. Especially those plants which grow on the flat surfaces of rocks, for instance on reefs, show a marked isodiametric development. But if the plant grows pendant from an overhanging rock, it develops the elongated type of frond.

Another condition, and dependent upon locality, is the movement of the water. Plants growing where they are continually exposed to the wash of the waves, back and forth, and from side to side, show far less marked longitudinal development than those which are exposed to the movement of the water in but one direction. This is very well illustrated by *P. naiadum* And. f. *major*. The author had an opportunity to observe this plant growing on *Zostera* in the lagoon at Bolinas, Marin County, California. The lagoon, which is long and narrow, was, in the summer of 1899, protected by a high bar, so that at the rise of the tide the water flowed in very regularly for a number of hours, till the turn of the tide, when it flowed out as regularly. The blades of *Zostera* and the fronds of *P. naiadum* were bent in the direction taken by the water and the latter showed a marked elongation, so great, indeed, that the writer felt entitled to consider them a special form of the species, since they were fully twice as long as the blades of *P. naiadum* growing on *Phyllospadix*, and since there existed some other minor differences as well. The latter form was consequently designated as *P. naiadum* f. *minor*.

An interesting instance of great length attained by exposure to the motion of waves in one direction was found in a specimen of *P. perforata* f. *lanceolata*, which grew on a rock buried in the sand of the gently sloping shore of the Presidio, San Francisco, California. This specimen attained a length of 325 centimeters, which to the author's knowledge is the greatest length ever attained by any specimen of this species of *Porphyra*. This extraordinary longitudinal development (the average length is but thirty to forty centimeters) was evidently due to the plants being stretched out at full length every time a wave rolled in or went out.

P. nereocystis, growing in three to five fathoms of water on the stipes of *Nereocystis lütkeana*, often attains a great length, specimens of over three meters in length having been collected at Monterey. A specimen of *P. variegata* collected at Santa Cruz by Dr. Anderson measured seventy-nine centimeters. On the other hand, we have found a fertile specimen of *P. perforata* but two centimeters long. But the plant which in its adult stage is the smallest of all Pacific Coast species of *Porphyra* is *P. naiadum*, which often bears fruit when but one centimeter long.

The width of the fronds also varies considerably. While the writer has measured specimens of *P. nereocystis* which were fully forty-nine centimeters in diameter, some mature specimens of *P. perforata* f. *lanceolata* collected by Dr. W. A. Setchell at Monterey, California, measured in their widest part but twenty-five hundredths of a centimeter.

In regard to the part age plays in the determination of the shape of the fronds, it must be said that while the younger plants as a rule possess the elongated type of frond, the older plants generally have a greater width. A microscopical examination reveals the fact that the divisions of the cells of the younger fronds are usually parallel to each other and at right angles to the longer axis of the frond. In the older plants, where the development is more isodiametric, we can readily recognize more or less isodiametric groups of cells, which evidently arose from a single cell. Of course, environmental conditions have much to do with this.

The base in *Porphyra* varies from cuneate to cordate, or even umbilicate, and is sometimes cucullate, as in *P. nereocystis*. These various forms of base depend just as much as size, etc., upon age, zone, and locality. An umbilicate base is found on the fronds of plants growing on flat surfaces (*P. laciniata* f. *umbilicalis*); one is liable to meet with a cordate base in the older fronds; while a cuneate base is found on the fronds of plants which grow in exposed places.

In the genus *Porphyra*, we may distinguish between two kinds of attachment, the one cushion-shaped and parenchymatous, the other discoid and rhizoidal. The latter form of base has been amply discussed and illustrated by Bornet and Thuret (1878), and it will suffice to say here that the cells in the immediate neighborhood of the base produce rhizoid-like projections, in thickness from two-tenths to one-tenth the diameter of the cell, which grow down through the jelly between the cells and the cuticle. That these hyphæ actually grow, and that the older the plant grows the more of these projections are produced, is demonstrated by treating the base of a young specimen of *Porphyra perforata* with Schultze's macerating fluid. This dissolves the jelly, and the weight of a cover-glass crushes the preparation sufficiently to show the details. From each of the thick-walled cells near the base, a hypha may be seen growing out, which may be longer or shorter, some being even but a few microns in length. Their course is more or less direct. Most of them extend down to the substratum. Consequently, though the frond about one centimeter above the base is normal, the part lower down is very much thickened by an ever increasing number of these projections, which finally form a dense network, in which it is impossible to trace the individual hyphæ. According to Agardh (1882), these hyphæ possess no septa, at least, he has been unable to see them; for he observes: "Hæc fila radicantia Porphyræ mihi semper inarticulata obvenerunt, * * * ." They are long, slender, tapering threads, averaging one to two microns in thickness. Of course, the longer they become, the less evident their tapering nature

is. In fact, the diameter of the larger hyphæ appears to be the same for the whole length. While the majority agree in this regard, if we follow them down to the base a difference soon becomes apparent. Some of the hyphæ come to an abrupt end, their diameter remaining constant, the contents remaining hyaline and parietal, and no septa being present; but others show a greater or lesser increase of thickness at the tip for a greater or lesser length, and a few even branch or at least show indications of branching (Pl. XX, figs. 7–10). In some of these, septa have been demonstrated. The swollen ends contain protoplasm. Whether these ends are to be considered as haustoria, and whether the hyphæ enter the cells or intercellular spaces of the host-plant, or whether they merely adhere to the substratum, are questions to which the author can give no definite reply. In sections of the base of a specimen of *Porphyra perforata* which grew on *Phyllospadix*, it was impossible to determine the course of the hyphæ. The same was true for plants growing on wood. Young specimens of *Porphyra perforata* growing on barnacles were treated with one per cent. nitric acid; but after dissolving the calcium salts, it was impossible, partly owing to the confusing mass of parasitic algæ which flourished in large numbers on and in the shells, but especially to the presence of chitin, to follow the hyphæ in their course.

The ever increasing number of hyphæ adds considerably to the thickness of the frond, the latter within one-half a centimeter of the disc often measuring two hundred microns or more, while the strength must be increased a hundred-fold. The cells which give rise to these hyphæ, especially those situated more towards the disc, are but imperfectly seen, even in a specimen not quite four centimeters long, being obscured by the hypha-like projections which surround them. It is easy to conceive that being thus partly excluded from the light these cells should undergo some change. They lose their purplish color, have yellow-brown cell-contents, and their walls are considerably thickened.

At first it seemed exceedingly improbable to the author that the cells to which the function of the attachment of the frond was delegated should finally produce fruit; but a knowledge of the fact that the formation of spores in these cells was observed by Bornet and Thuret (1878) led him to make a more careful study of the basal cells. Up to this time, however, the author has been unable to demonstrate a single cell which both emitted a hyphal thread and bore fruit, though the oldest obtainable specimens were investigated. It must therefore be concluded that if ever the contents of these cells are transformed into spores, this must be but rarely the case.

The areolate, lighter colored portion of the frond, about one centimeter in diameter, directly surrounding the attachment found in all specimens of *Porphyra* as known to the writer, with the exception of *P. naiadum*, probably finds its reason in two causes. The first is the partial loss of color of the cells near the base, caused by decreased activity owing to the large number of rhizoid-like projections which separate them from the surface of the frond. The other cause may be looked for in the large number of rhizoid-like projections with hyaline walls, resulting in an increased thickness and consequently increased density of the lowest part of the frond.

Some of the species of *Porphyra* are slightly stipitate. On this coast only *P. leucosticta* shows this to any marked extent. The stipes appear to possess the same structure as the discs.

A cushion-shaped base is, as far as the writer is aware, found in but a single species of *Porphyra*, viz., *P. naiadum* And., a species peculiar to the Pacific Coast. *P. naiadum* has been found growing on eel-grass, either on *Phyllospadix* in exposed places in the sublitoral zone or on *Zostera*, sheltered, in lagoons.

On examining the blades of *Phyllospadix* during the winter months, we find here and there small reddish brown, cushion-shaped growths, which to the superficial gaze appear like colonies of diatoms. Continued observation shows a gradual increase in the number of these wart-like,

more or less flattened structures. Finally, they cover the blades of eel-grass in such large numbers that they grow next to and over each other, and lose their natural hemispherical shape, obscuring the normal color of the eel-grass, and giving a rough appearance to the blade.

As the season advances, examination with a lens shows a greater or smaller number of short, blunt protuberances issuing from the wart-like growths. Under the microscope they appear to be composed of a number of cells placed end to end. Further observation demonstrates the fact that these cells, by division in two planes, give rise to a monostromatic frond. From this it is but a step to establish a genetic connection between the hemispherical structures on eel-grass and the fully grown fronds of *P. naiadum* on the same host-plant. Evidently we have the prothalloid form of *P. naiadum* before us. This was already suspected by Dr. Setchell when he called my attention to the matter.

The prothallium, when young, consists of but a single layer of cells, placed side by side on the blade of the eel-grass. For a certain length of time these cells continue to divide in a single plane. After that, division in the second plane begins to take place, gradually giving rise to the wart-like growths referred to above. In section they appear to consist of layers of large, thin-walled, parenchymatous cells (Pl. XXI, fig. 19). The cells of the central layers possess ordinary cell-contents, but only a very small chromatophore. The two or three outer layers are made up of slightly smaller cells, and possess a large chromatophore. The cells of the layer adjacent to the surface of the blade of the host-plant also contain a large chromatophore. Upon these latter cells evidently devolves the function of attaching the prothallium to the eel-grass. Each cell is extended so as to form a short, sharp, unicellular rhizoid. The writer has been unable to determine with any satisfaction whether these rhizoids entered the cells of the host-plant or not. Careful sectioning and staining has failed to reveal anything of the kind. But in material which had been shrunken by reagents, only the rhizoids at the periphery of the cushion-shaped base were attached to the eel-grass, the

central part of the base having shrunk away, exposing the rhizoids. From this it would appear as if the rhizoids entered, to a slight extent only, the cuticula of the host-plant.

The young fronds arise from the cells of the external layer of the prothallium. Evidently any cell may give rise to a frond by a division in one plane, in advance of the surrounding cells which form part of the external layer of the cushion-shaped base. When in this manner a filament of some five or six cells has been formed, the cells of the filament begin to divide in two planes, thus giving rise to a membranous frond, the length of which when fully grown seldom exceeds six centimeters.

The number of fronds a prothallium may give rise to appears to be indefinite, every cell of the outer layer of the base seemingly being capable of producing a frond. A frond may be formed during the first stage of the existence of the prothallium, cases having been observed where frond formation evidently took place when the prothallium was but two cells in thickness. The formation of a frond by a cell of the outer layer does not mean cessation of growth for the other cells of the outer layer, since frequently a frond may be found, the base of which lies in a depression of the prothallium several cells deep.

An attempt was made to ascertain, if possible, if when the frond has reached a certain size, the cells of the frond, in the neighborhood of the base, produced rhizoid-like projections such as are found in the corresponding cells of other species of *Porphyra*. But an examination of the bases of a large number of mature fronds of *P. naiadum* failed to reveal these structures.

When *Porphyra naiadum* occurs on *Zostera*, it produces the same wart-like growths, but only on the extreme margins, not on any part of the surface of the blade, and they appear to be smaller.

Other species of *Porphyra* probably occur but seldom on eel-grass. The only species found by the writer to occur occasionally on the same host-plant were *P. perforata*, *P. laciniata*, and *P. abyssicola*.

The value of the prothalloid base lies evidently in the power to form a large number of fronds rapidly, which in the production of new fronds in case of accident is clearly of great importance.

As far as the author is aware, *P. naiadum* has never been found growing on any substratum other than eel-grass, nor has a cushion-shaped base such as here described ever been found in any other species of *Porphyra*. The nearest approach to such a description is that of the base of *P. coccinea* J. Ag., such as is found in Agardh's "Till Algernes Systematik" VI (1882). But judging from this account, the base is hollow, being formed by the involution of the edges of the young frond. When the frond grows older the base finally flattens out.

It is worthy of note, that in connection with *P. coccinea* Agardh mentions a *Porphyra* occurring in large numbers on the Pacific Coast; it grows on seaweeds (!), and judging from the description given might possibly be *P. naiadum*.

The color of the fronds of the different species of *Porphyra* is such as to lead one to place the genus among the Florideæ. But the color is far from being constant. A hundred different shades may be met with, for even the color of the different fronds belonging to one species varies; so that an attempt to describe a species by the color would be futile. While the color of one species (*P. tenuissima*) is, as a rule, a delicate pink, others are a bright red or even crimson, as *P. abyssicola*. *P. laciniata* exhibits a decidedly purple color, while *P. perforata* appears mostly yellow-brown. The frond of *P. variegata* is crimson when sterile, while when fruiting it acquires the beautiful variegated appearance indicated by its name.

An important fact is that the color of herbarium specimens generally changes. This was most notable in *P. perforata*. Specimens which when collected had a yellow-brown tint generally became a deep blue-purple. Some of the fronds of *P. nereocystis* underwent a change in the herbarium, while others retained the original dull brown-red color.

Especially did specimens which were rough-dried and afterwards soaked in fresh or salt water for mounting

purposes seem to change color on drying. A similar change was noted in sheets of Asakusa Nori (see Economic Uses.) which were of a yellow-brown color when bought in San Francisco, but having been kept for several months in a closed paper box turned purple. A sheet of Asakusa Nori which had accidently been left partially exposed to the air and light for about a month, showed after that time a brilliant violet coloring in the exposed portion, while that part of the sheet which was not exposed retained its original yellow-brown tint.

As far as can be judged from the statements of various authors, as well as from our own observations, it appears that the color of certain species varies according to the locality. This is well illustrated by *P. leucosticta* Thur. It seems that the European specimens of *P. leucosticta* are of a distinctly yellow color when fresh, and when dried a delicate purple-pink tint. But the specimens of *P. leucosticta* found on the Pacific Coast, if gathered early in the season, are deep-pink, becoming lighter as the season advances.

From the above, it will be seen that specimens of *Porphyra* should, whenever possible, be mounted fresh; that even then the color is of small value from a systematic point of view; and that it is most undesirable, in fact, impracticable, to use the color of a frond as the criterion for the species, though it is often of great value in indicating its position.

It remained for J. G. Agardh to call attention to the monostromatic and distromatic nature of the fronds of the different species of *Porphyra*. These characters have been found to be absolutely constant in all species, with the exception of those belonging to what we may call the "*miniata*" group, which includes besides *P. miniata*, *P. amplissima*, *P. tenuissima*, and *P. abyssicola*. The first three species are, as a rule, distromatic, though places may be found which exhibit a monostromatic character, especially towards the edges. Fronds of *P. abyssicola*, which species was first described by Kjellman as monostromatic, have been found by Rosenvinge and by the author to sometimes exhibit a distromatic character, either through the whole frond or in portions of it.

Kjellman applies the name *Diploderma* to all distromatic species, but the above mentioned results lead the writer to agree with Rosenvinge in applying the name *Porphyra* to all members of the genus, while retaining *Diploderma* (*Wildemania* de Toni) as a subgeneric name for the distromatic fronds, the more so as in habit and external character the monostromatic and distromatic species agree in all respects.

Two sources of confusion in determining the number of layers in the frond exist. The first is the age of the frond, the second, the formation of reproductive cells. Young fronds of distromatic species are frequently monostromatic. This monostromatic character persists in the vegetative portion of the frond even after the fruit has been formed (*P. abyssicola*) (Rosenvinge, 1893). The same results were obtained by the author in his observations on *P. abyssicola*, but he cannot confirm Rosenvinge's statement, that in the distromatic forms the inferior portion of the thallus is composed of a single layer of cells. "Dans les formes distromatiques, du reste, la partie inférieure du thalle est composée d'une seule assise de cellules" (Rosenvinge, 1893, p. 84). On the contrary, if we do not consider those of *P. abyssicola*, but few, if any, distromatic specimens were found which were monostromatic at the base. As a rule, whenever a frond of a distromatic species was partly monostromatic, the monostromatic portion was found in the region of the tip at the edge.

Monostromatic species practically become distromatic as soon as the reproductive bodies begin to form. The second division of either the antheridium or sporocarp-mother-cells takes place in a direction parallel to the surface of the frond, and in this manner gives rise to two layers of cells which are often difficult to distinguish from purely vegetative cells; so that in deciding their nature, the cells of the surrounding tissue must be taken into account.

The thickness of the fronds of the various species of *Porphyra* is more or less variable, and though not an absolutely specific character, taken in conjunction with other

characteristics it is of great value, especially in indicating the position of a sterile frond.

As a rule, the thickness of the fertile part of the frond is much greater than that of the sterile part. This seems to be due to the swelling of the jelly surrounding the reproductive bodies at the time of ripening. Also, wherever we have a diœcious frond, or where the antheridia and sporocarps are born on separate portions of the plant, the thickness of the antheridial portion is greater than that of the sporocarpic portion. This would bear out our hypothesis that increased thickness is due to the swelling of the jelly, since there exists a larger number of partitions consisting of jelly between the antherozoids than between the carpospores.

P. variegata exhibits a very marked thickness and stratification of the jelly-walls surrounding the vegetative cell. The walls of the vegetative cells of all fronds of this species examined were of this nature; consequently the author feels entitled to consider this a diagnostic character, the more so as the only other *Porphyra* which possesses much thickened cell-walls differs widely from *P. variegata* in habit and external characters.

The plant here referred to is *P. perforata* f. *segregata*. The walls not only of the vegetative cells but also of the reproductive cells are much thickened, especially those produced by the first reproductive division of the antheridium. In fact, the upper and lower groups of antherozoids are noticeably separated, which lends the cross-section of the antheridial portion of the frond a most characteristic appearance.

The outer jelly-walls of nearly all the fronds examined were infested with bacteria which formed narrow lines perpendicular to the surface of the frond, reminding one of the canals formed by the "spermatium" at the time of the fertilization of the "procarp", as described by Berthold (1882). These "canals" were found in the jelly surrounding the vegetative and antheridial cells, as well as in that surrounding the sporocarps.

In regard to the shape of the vegetative cells, it may be said that while in the monostromatic fronds the cells are

either cubical or more frequently higher than broad, the vegetative cells of the fronds of the distromatic species vary, as a rule, from cubical to broader than high. Exceptions to this rule are found in *P. nereocystis*, a monostromatic species which sometimes possesses cells which are broader than high, and in *P. variegata*. While the vegetative cells of the younger sterile fronds of the latter species are usually square, the vegetative cells of the older fertile fronds are much higher than broad and often have a fusiform appearance. Judging from the fact that the vegetative cells are found between the reproductive cells, it is suggested that the shape of the former is due to pressure exerted by the reproductive cells, which before dividing gorge themselves with protoplasm, and when fully ripe swell to an abnormal size, owing to the partial dissolution of the jelly partitions separating the individual spores.

During the study of the species of *Porphyra* of the Pacific Coast, the fact gradually made itself felt that the reproductive bodies are of the greatest diagnostic value, and that habitat, color, and thickness of frond can only be used to determine species in connection with the number of divisions of the antheridia and sporocarps.

Since the object of this paper is merely to give a systematic account of the species of *Porphyra* of western North America, and it is not designed to throw light on the sexuality or nonsexuality of the genus *Porphyra*, the author uses the terms sporocarp and antheridium merely to indicate the larger and smaller bodies, which by some are believed to play a part in sexual reproduction, without necessarily ascribing a sexual character to these bodies. The same is true for the asexuality of the monospores. But it must be said that in no case even the slightest indication of sexuality has been observed, though many sections of sporocarps in all stages of development were examined. Neither has the author been able to observe an amœboid movement of the liberated carpospores, nor flagelliform appendages to the bodies contained in the antheridia, nor any movement on the part of these bodies, though observations were made to

determine this point if possible. The results obtained by the writer therefore agree rather with those obtained by Reinke and Bornet than with those of Berthold and others.

While some species of *Porphyra* are monœcious, others are diœcious. In some cases fronds have been found which evidently approached closely to those of a monœcious species, but which differed from it in being diœcious and in exhibiting some slight differences of habit, etc. Such fronds have been referred to the original species but were separated from it under a form-name.

We may distinguish two forms of monœcious fronds. In the one, antheridia and sporocarps are separated in patches. These patches are usually sharply defined, especially at the edges of the frond, owing to the lighter color of the ripe antheridia and to the more intense color of the ripe sporocarps. This arrangement is usually met with in the fronds of the monostromatic species. In the other form, the antheridia and sporocarps occur side by side, so that the frond has a uniform color. This occurs in the distromatic species, and more particularly in what the writer has found convenient to designate the "*miniata* group," which includes *P. amplissima, P. miniata, P. tenuissima and P. abyssicola*.

Vegetative cells are frequently found mixed with the sporocarps, among both monostromatic and distromatic species, and in larger or smaller patches. While the distromatic species show this constantly, the monostromatic species often fail to show these vegetative cells among the sporocarps. Hardly ever has the author found any vegetative cells mixed in with the antheridia of the monostromatic species.

Among the sporocarps there appear frequently bodies which by various authors have been called monospores. They seem to be formed by the arrest of division in one of the segments of the sporocarp or by one of the vegetative cells lying among the sporocarps. What differentiates them from the vegetative cell proper is a greater thickness of the cell-wall and a larger amount of protoplasm. The chromatophore may be seen lying in or near the center of the cell.

These monospores can easily be distinguished from the dead vegetative cells lying among the sporocarps. The dead cells possess likewise a thick wall but apparently contain yellowish, homogeneous, highly refractive cell-contents, in which no chromatophore can be discerned.

Whether monospores are sexual or not, or whether they possess any reproductive power, the author has, notwithstanding a series of careful experiments, been unable to determine.

The reproductive bodies are usually first formed at the margins and gradually spread over the whole frond. Bornet even observed spores in the basal cells of *P. laciniata*. Under the microscope we can trace the various stages of division from the original vegetative cell to the fully ripe sporocarp. This is especially easy in the species where antheridia and sporocarps occur side by side in patches. Observation shows that each vegetative cell gives rise to a single sporocarp. The sporocarp by two more or less simultaneous divisions at right angles to each other and to the surface of the frond finally consists of four segments. In some species division proceeds no farther, and four carpospores are the result; but in other species, where the fully ripe sporocarp contains more than four carpospores, the cruciate division is followed by a division parallel to the surface of the frond, giving rise to eight segments, which by further cruciate division perpendicular to the surface of the frond in each of the resulting cells may give rise to thirty-two carpospores.[1]

In the formation of the antheridia, starting from the vegetative cell equivalent to the mother-cell of a sporocarp, there first takes place a vegetative cruciate division perpendicular to the surface of the frond, which gives rise to four antheridial cells. Consequently, owing to this additional vegetative

[1] The author understands under "cruciate" division, two divisions in different directions, at right angles to each other and to the surface of the frond, and which are simultaneous or nearly so. Under "transverse" or "parallel" division the writer understands a division parallel to the surface of the frond. Though these divisions are, of course, but seldom if ever strictly parallel or at right angles, the use of the terms "cruciate" and "parallel" is a great convenience, doing away with a lengthy explanation.

division of the antheridium-mother-cell each sporocarp corresponds to four antheridia.

The author distinguishes between a vegetative division and a reproductive division by the aid of the partition walls laid down by these divisions. A jelly-wall formed after a vegetative division is, as a rule, much thicker than one formed after a reproductive division. Furthermore, a wall of the latter kind dissolves when the frond is fully ripe, so that the reproductive bodies become arranged more or less irregularly; something which is very clearly shown in the antheridia of *P. leucosticta*.

The first reproductive division of the antheridium is parallel to the surface of the frond, corresponding to the first transverse division of the sporocarp, and is followed by a cruciate division in both segments. In fact, parallel and cruciate divisions alternate until the number of antherozoids peculiar to the species has been formed. The only difference, therefore, between antheridia and sporocarps lies, if we do not consider their origin, in the larger number of divisions which the former undergo.

Though the manner of division of antheridia and sporocarps is fairly constant, yet a large number of variations take place. The most frequent among these is the direction of the last division in either antheridia or sporocarps, which is not necessarily parallel or perpendicular to the surface of the frond, but is often oblique, and is occasionally omitted altogether in some of the segments of the sporocarp or antheridium. Rarely an additional division takes place in some or all of the segments of the sporocarp or antheridium.

Cases have been met with, where the vegetative division of the antheridium-mother-cell is not as evident as usual, and the whole vegetative cell apparently becomes an antheridium, so that four times the usual number of antherozoids are formed.

Occasionally the first cruciate division of the sporocarp-mother-cell is vegetative instead of reproductive, and only one-fourth the usual number of carpospores are found.

With the material at the disposal of the author, he has been able to distinguish between four types of division of the reproductive bodies, the differentiation into types being based upon the number of antherozoids and carpospores produced. The first is the *Porphyra perforata* type. Here thirty-two carpospores are produced, the sporocarp undergoing first a cruciate division, followed by a parallel division in each of the segments, which is again followed by a cruciate division of each segment. If we represent a vegetative cell by a cube, and indicate the two horizontal lines respectively as a and b, and the perpendicular as c, we can, by the aid of the formula $32\left(\frac{a}{4}, \frac{b}{4}, \frac{c}{2}\right)$ readily form a diagram such as is represented in fig. 25, which shows the manner of division.

The antherozoids contained in each antheridium number, in the *P. perforata* type, 128, and are formed by alternating parallel and cruciate divisions, the first division of the antheridium being parallel to the surface of the frond.[1] The manner of division may be represented by the formula $128\left(\frac{\frac{1}{2}a}{4}, \frac{\frac{1}{2}b}{4}, \frac{c}{8}\right)$ (fig. 28). To this type belong *P. perforata*, *P. perforata* f. *lanceolata*, *P. perforata* f. *segregata* and *P. nereocystis*.

The second is the *P. leucosticta* type. Here eight carpospores arise from a cruciate division of the sporocarp-mother-cell, followed by a parallel division of the four segments. This may be represented by the formula $8\left(\frac{a}{2}, \frac{b}{2}, \frac{c}{2}\right)$ (fig. 24).

The antherozoids contained in each antheridium of the fronds of species belonging to the *P. leucosticta* type number sixty-four. They are formed by first a parallel division of the antheridium, followed by a cruciate division, after which a second parallel and a second cruciate division take

[1] It must be remembered that an antheridium is but one-fourth as large as a sporocarp, the first division of the antheridium-mother-cell, which corresponds to both a vegetative cell and a sporocarp-mother-cell, being vegetative and cruciate.

place in each segment, so that the arrangement of the antherozoids is represented by the formula $64\left(\frac{\frac{1}{2}a}{4}, \frac{\frac{1}{2}b}{4}, \frac{c}{4}\right)$ (fig. 27). To this type belong *P. leucosticata*, *P. laciniata*, and it may be predicted, judging from the carpospores, that *P. naiadum* also belongs to this section, the antheridia in this species being as yet undetected.

Among the distromatic forms the remainder of the types are found. First we have the *P. amplissima* type, with eight carpospores and sixteen antherozoids, and the formulæ $8\left(\frac{a}{2}, \frac{b}{2}, \frac{c}{2}\right)$ (fig. 24) and $16\left(\frac{\frac{1}{2}a}{2}, \frac{\frac{1}{2}b}{2}, \frac{c}{4}\right)$ (fig. 26); then the *P. miniata* type, with four carpospores and eight antherozoids, and the formulæ $4\left(\frac{a}{2}, \frac{b}{2}, c\right)$ (fig. 23) and $8\left(\frac{\frac{1}{2}a}{2}, \frac{\frac{1}{2}b}{2}, \frac{c}{2}\right)$ (fig. 25). To this belong *P. miniata*, *P. tenuissima* and *P. abyssicola*.

Of the two other distromatic species which occur on the Pacific Coast, but one form of fruit has been found, so that they can hardly be brought forward as types.

III. DISTRIBUTION.

It is almost impossible to obtain a correct idea of the distribution of the older species of *Porphyra*, as frequently the name *P. laciniata* was applied to various species which since have been separated from it. However, the author believes that it may be said with some degree of certainty that *P. laciniata* occurs on the western shores of Europe, from the Norwegian Polar Sea (71° N. lat.) to the Mediterranean (40° N. lat.) and on the Atlantic coast of North America from Greenland (67° N. lat.) to New Jersey (40° N. lat.). It has never been authoritatively reported from the eastern shores of Asia; for though older authors have mentioned it, yet *P. laciniata* was not included by Kjellman (1897) among the Japanese species, and he even expresses some doubt as to its occurrence. On the Pacific Coast of North

America *P. laciniata* has been reported from Orca, Alaska, from Yakutat, and from Amaknak Island (between 61° and 54° N. lat.). *P. laciniata* f. *umbilicalis* was reported by Professor Setchell (1899) from the Pribilof Islands.

Porphyra leucosticta Thur. does not appear to possess such a wide range as the species just discussed. It is found on the Atlantic coast of England, Germany, and France, and appears to be abundant in the Mediterranean. Collins (1884) and Holden (1897) have detected it on the eastern shores of North America, and while it may have a wide distribution on the Pacific Coast, it has as yet been reported from but a single locality, Monterey Bay, California (36° 45′ N. lat.).

Up to the present time the members of the "*miniata*" group, under which the author includes *P. amplissima*, *P. miniata*, *P. tenuissima*, and *P. abyssicola*, have been reported by European collectors only from the more northern latitudes (60°–80° N. lat.). On the west coast of North America these species have not been reported from so far north, ranging between 36° 45′ and 60° N. lat.

P. amplissima was first found by Kjellman (1883) in the Norwegian Polar Sea, and it has since been reported from both the east and west coast of Greenland. Kjellman did not detect this species in any of the localities visited by the Vega Expedition. Since then it has been collected at Orca, Alaska (60° 30′ N. lat.), and near Coupeville, Washington (48° 10′ N. lat.).

P. tenuissima occurs on the shores of Norway, Iceland, and Greenland. It has never been found in the Bering Sea, but it has been collected at Sitka, Alaska (57° N. lat.).

P. miniata is met with on the coasts of Norway, and attains its greatest latitude on the northwest coast of Spitzbergen (79° 49′ N. lat.). It is also found on the east coast of Norway and in Baffin Bay, whence it descends to New Foundland.

There exists some doubt in the author's mind as to the occurrence of the typical *P. miniata* on the Pacific Coast. Only an extensive collection of specimens gathered on

numerous expeditions along the coast can lead to a definite conclusion. But the occurrence of a variety designated by us as *P. miniata* f. *cuneiformis* is an undoubted fact. It has been collected in the Gulf of Alaska (60° N. lat.), at Coupeville, Washington, and as far south as Monterey Bay, California (36° 45′ N. lat.).

P. abyssicola is reported by Kjellman and others from the north coast of Norway and Russia and from Greenland. What the author believes to be *P. abyssicola* has been collected on the Pacific Coast at Whidby Island, Washington (48° 10′ N. lat.).

Five species of *Porphyra* appear to be peculiar to the Pacific Coast. In some cases it is possible to ascribe a reason for this. *P. nereocystis*, which as far as can be judged selects *Nereocystis lütkeana* exclusively as its host-plant, is necessarily limited to the region of distribution of this species of *Nereocystis*. It has been reported from St. Paul, Kadiak Island (57° 30′ N. lat.), from Coupeville, Washington, and from the Californian shores (33°40′ N.lat.).

P. naiadum, growing on *Zostera* and *Phyllospadix*, seems limited to the Pacific Coast. Though other species of *Porphyra* occur on *Zostera*, both in Europe and on American shores, yet no case is known to the author where *P. naiadum* was found growing on eel-grass in waters other than those of the Pacific, where it extends from Coupeville, Washington (48° 10′ N. lat.) to San Diego, California (32° 20′ N. lat.).

P. perforata, so closely allied to *P. laciniata*, attains nearly the same northern latitude as the latter species, but extends far lower down the Pacific Coast. Of the two varieties of this species the author has been able to find only one, *P. perforata* f. *lanceolata*, at San Francisco and at Monterey, while the other occurs from Washington to Mexico (San Roque) (47° 30′–27° 8½′ N. lat.).

P. variegata was first found by Kjellman at Bering Island (Vega Expedition). Since then it has been reported by various collectors along the Pacific Coast, from Whidby Island, Washington, to San Pedro, California (48° 10′–33° 40′ N. lat.).

P. occidentalis has been found in but a single locality, Monterey Bay, California (36° 45′ N. lat.).

DISTRIBUTION OF THE PACIFIC COAST SPECIES OF PORPHYRA.

P. occidentalis.	*P. variegata.*	*P. abyssicola.*	*P. tenuissima.*	*P. miniata.*	*P. amplissima.*	*P. naiadum.*	*P. nereocystis.*	*P. perforata.*	*P. leucosticta.*	*P. laciniata.*	West Coast of North America.
	*				*						Behring Sea.
	*		*	*	*		*	*		*	Gulf of Alaska.
	*	*		*	*	*	*	*			Vancouver, Whidby Island, Seattle.
						*	*	*			San Francisco.
*	*			*		*	*	*	*		Monterey Bay.
								*			Santa Barbara.
	*					*	?	*			San Pedro, San Diego.
				*					*	*	New England.
									*	*	West Europe.
									*	*	Mediterranean.
		*	*	*	*					*	Greenland.
		*	*	*	*					*	Arctic Ocean and Spitzbergen.

IV. DESCRIPTIONS OF SPECIES.

KEY TO THE PACIFIC COAST SPECIES OF PORPHYRA.

1. Fronds monostromatic 2
   Fronds distromatic 7
   Fronds monostromatic or distromatic 11
2. Base cushion-shaped, consisting of parenchymatous cells ... *P. naiadum*
   Base discoid, consisting of agglutinated, rhizoid-like cells 3
3. Thirty-two spores in each sporocarp 4
   Eight spores in each sporocarp 6
4. Fronds monœcious 5
   Fronds diœcious ..... *P. perforata* f. *lanceolata*
5. Fronds brown-purple, 45–150μ thick ..... *P. perforata*
   Fronds brown-purple, 60μ thick, with thick partition walls ..... *P. perforata* f. *segregata*
   Fronds red-purple, 25–60μ thick, with thin partition walls .. *P. nereocystis*
6. Fronds monœcious ..... *P. leucosticta*
   Fronds diœcious ..... *P. laciniata*

7. Fronds monœcious 8
   Fronds apparently dicœcious 10
8. Sixteen antherozoids in each antheridium ..... *P. amplissima*
   Eight antherozoids in each antheridium 9
9. Fronds 25μ thick, except at base ..... *P. tenuissima*
   Fronds 30–75μ thick ..... *P. miniata* f. *cuneiformis*
10. Fronds 100–220μ thick ..... *P. variegata*
    Fronds 45–75μ thick ..... *P. occidentalis*
11. Fronds 25μ thick ..... *P. abyssicola*

1. Porphyra laciniata *(Lightf.) Ag.*

Systema Algarum, p. 190, 1824.

Porphyra laciniata (LIGHTF.) C. A. AGARDH, Systema Algarum, p. 190, 1824; Icones Algarum Europæarum. Tab. XXVII, 1828. HARVEY, British Marine Algæ, p. 216, 1849. JANCZEWSKI, Etudes Anat. sur les Porphyra, p. 352, 1873. BORNET et THURET, Etudes Phycologiques, p. 58, 1878. J. G. AGARDH, Till Algern. Systematik., VI, p. 67, 1882. FARLOW, Marine Algæ of New England, p. 111, 1881. HUS, Zoe, Vol. V, p. 62, 1900.
Ulva laciniata AGARDH, Species Algarum, p. 404, 1822.
Porphyra vulgaris HARVEY, Phyc. Brit., Pl. CCXI. fig. 1, 1851.
Wildemania ? laciniata DE TONI, Sylloge Algarum, Vol. IV, p. 20, 1897.

Fronds membranous, 10–80 cm. long, 5–30 cm. broad, linear when young, becoming lanceolate or broadly expanded and much laciniate when older; base obtuse to cordate; sessile on disc; gray-purple; monostromatic, vegetative part of frond 30–45μ thick, cells square with rounded angles or higher than broad, surface jelly 7–8μ thick; dicœcious, sometimes monœcious, sporocarps and antheridia forming a marginal zone, 8 (or 16) carpospores in each sporocarp, 64 (or 128) antherozoids in each antheridium.

The author has, up to this time, been unable to find any data in regard to the size this plant attains, but has received the impression from the writings of others, as well as from an examination of specimens, both American and European, that 30 centimeters is the average length attained by the fronds of this species, which suspicion was confirmed by the study of a number of specimens of our western coast. But recently there have come to our notice two specimens from *Orca*, Alaska, one of which measured 60 centimeters, while the other was 80 centimeters long, with a breadth of 30 centimeters. But such specimens do not indicate the size the species normally attains; a length of 30 centimeters and a breadth of 10–15 centimeters represent the average measurement.

The shape of the fronds is far from being constant, and appears, as in other species of *Porphyra*, to be strongly influenced by environmental conditions. While the young frond is liable to be more or less linear, even up to the time when it attains a length of from 15 to 20 centimeters, most specimens of that size show a considerable lateral development. Older specimens are liable to be much laciniate.

The form of the base, like the shape of the frond, depends on the surroundings; some specimens, probably those which were exposed to the motion of the waves, exhibiting an obtuse base when young, and a more or less cordate base when older.

The color of the fronds is fairly constant, being as a rule an even gray-purple, which increases in intensity with age. Various shades may be met with.

The frond is proliferous and deeply folded and at times beautifully crenulate.

Attachment is by a disc. The aureole around this disc, so pronounced in *P. perforata*, is here but slightly marked.

The vegetative portion of the frond is uniform in thickness, measuring from 30–45 microns. The cells are square or from two to three times as high as broad. The angles are always rounded. There is but little jelly between the cells, and the outer layer of jelly is not very thick, though this varies more or less owing to different conditions of exposure to air and heat, measuring about 7 or 8 microns.

The fronds are strictly monostromatic in the vegetative part.

Porphyra laciniata is as a rule dicœcious, though some instances were found when the fronds were monœcious. The fruit occupies a marginal zone. When antheridia and sporocarps are found on the same frond, they occur in patches very much as in *P. perforata*. The writer has never met an instance where a few sporocarps were intermixed with a large number of antheridia, such as are found in the *miniata* group; but a larger or smaller number of vegetative cells and monospores may be found among the sporocarps. Nor has he ever found vegetative cells among the antheridia.

Sporocarps.—The reproductive bodies of *P. laciniata* belong to the *P. leucosticta* type. Each sporocarp contains eight carpospores, which arise from the vegetative cell by a cruciate division, followed by a parallel division, thus giving rise to eight carpospores in two tiers of four each. This appears to be the normal number, but frequently this mode of division undergoes changes Either the vegetative division proceeds one step further and only a parallel division takes place, thus giving rise to but two spores, or in the latter case an additional parallel division may occur, so that the sporocarp contains four carpospores in four tiers of one each. Again it is possible that after the normal cruciate and parallel divisions have taken place, another more or less complete division, usually parallel or oblique, forms sixteen or less carpospores.

Antheridia.—The antheridium-mother-cell first undergoes a cruciate division, giving rise to four antheridia. Each antheridium now undergoes its first reproductive division, parallel to the surface of the frond. This division is followed by a cruciate division and by a division parallel to the surface of the frond, in all segments, thus giving rise to sixteen bodies in four tiers of four each. These bodies divide by a cruciate division, giving rise to 64 antherozoids. This division is in many cases followed by a division parallel to the surface of the frond, so that each antheridium now contains 128 antherozoids arranged in eight tiers of 16 each.

The drawings of *P. laciniata* of Bornet and Thuret (1878) admirably illustrate these points, as well as those cases where the vegetative division of the antheridium-mother-cell is less apparent; so that we find 256 or 512 antherozoids in an antheridium, or where the vegetative division of the antheridium-mother-cell goes one step farther so that each antheridium contains but 32 antherozoids.

Economic Use.—According to the Rev. Albin Johnson, the Indians of Yakutat, Alaska, collect, cook and eat this plant.

Habitat.—On rocks or epiphytic on *Fucus evanescens*. Throughout the litoral and lower litoral zones.

Distribution.—P. laciniata appears to be limited on the Pacific Coast to the shores of Alaska (61°–54° N. lat.).

Localities.—Orca, Alaska (W. A. Setchell, No. 5164!); Yakutat, Alaska (Rev. Albin Johnson, No. 14!); Uyak Bay, Kadiak Island, Alaska (W. A. Setchell, No. 5099!); Sitka, Alaska (de A. Saunders, No. 136!); Annette Island, Alaska (de A. Saunders, No. 26!); Amaknak Island, Alaska (W. A. Setchell, No. 3269!, No. 3270!).

2. Porphyra laciniata f. umbilicalis *Ag.*

Icones Algarum, Tab. XXVI, 1828.

Porphyra laciniata f. *umbilicalis* C. A. AGARDH, Icones Algarum, Tab. 26, 1828. SETCHELL, Algæ of the Pribilof Islands. Fur and Fur-seal Islands of the North Pacific Ocean, 1899, p. 593. HUS, Zoe, Vol. V, 1900, p. 62.

Distribution.—Pribilof Islands, Bering Sea; U. S. S. Albatross (according to Setchell, 1899) (57° N. lat.).

3. Porphyra leucosticta *Thur.*

PLATE XX, FIGS. 1*a*-3*b*.

In le Jolis, Liste des Algues Marines de Cherbourg, 1864, p. 100.

Porphyra leucosticta THURET in LE JOLIS, Liste des Algues Marines de Cherbourg, 1864, p. 100. DE JANCZEWSKI, Etudes Anat. sur les Porphyra, 1873, p. 241. COLLINS, Bull. Torrey Bot. Club, Vol. XI, 1884, p. 131. FARLOW, Marine Algæ of New England, 1881, p. 112. HOLDEN, in COLLINS, HOLDEN & SETCHELL, Phyc. Bor.-Amer., Fascl. VIII, 1897, No. 376. HAUCK, in HAUCK & RICHTER, Phyc. Universalis, Fascl. IX, 1891, No. 401. HUS, Zoe, Vol. V, 1900, p. 63.

Porphyra atropurpurea DE TONI, Syll. Alg., Vol. IV, 1897, p. 17.

Membranous, 7–70 cm. long, 2–25 cm. broad, oblong with slightly undulate margin; base cordate, stipitate; color light pink; monostromatic, vegetative part of frond 25–50μ thick, cells once and a half to twice as high as broad, surface jelly thin; fronds monœcious, antheridia forming small, elongated, colorless patches among the dark-colored sporocarps; fruit marginal, gradually spreading over the whole frond, no vegetative cells intermixed with the reproductive cells, sporocarpic part of frond 25–50μ thick, eight carpospores in each sporocarp; antheridial part of frond 30–50μ thick, 64 antherozoids in each antheridium.

Porphyra leucosticta Thuret is an annual, only to be found in the spring months. It has never, to the writer's

knowledge, been collected on the Pacific Coast north of Monterey Bay. It appears to be exceedingly delicate, especially those parts of the frond which bear fruit. When examining an herbarium specimen, the method of soaking the desired portion in water, as pursued with *P. perforata* and others, is entirely inadequate, water causing the almost instantaneous dissolution of the jelly. It becomes therefore necessary to use a concentrated solution of corrosive sublimate.

P. leucosticta is fairly constant in shape, irrespective of size, varying from oval in the young plants to oblong in the older ones. It is but seldom laciniate, and the margin is but slightly undulate. The base is decidedly stipitate, something in which the western specimen differs from the eastern and European plants, which are at the most substipitate. Another difference between eastern and European specimens of *P. leucosticta* on the one hand, and the western plants on the other, lies in the color, which in the latter varies from cerise to dull brown, while the former appear much lighter in color.

This plant is, in the vegetative portion of the frond, constantly monostromatic. No indications of a distromatic nature have ever been found.

The fronds are monœcious. At first the fruit is found only at the tip and along the margins. In a ripe frond we find a colorless margin, consisting of antheridia, together with an empty network formed by the jelly-walls of those sporocarps and antheridia which have discharged their contents. Inside this margin, the sporocarps and antherida are intermixed, the antheridia usually forming irregular, elongated, colorless patches among the dark cerise sporocarps.

The thickness of the reproductive portion of the frond does not differ materially from that of the vegetative portion, measuring from 25–50μ. This is an additional reason for ascribing the increase in thickness in the reproductive portions of the fronds of other species of *Porphyra* to the swelling of the jelly, since in *P. leucosticta* Thur. there is

but a very small amount of jelly surrounding the cells, and it could not swell enough to make an appreciable difference in the thickness of the frond.

Vegetative cells are but rarely found intermixed with the reproductive cells.

Sporocarps.—Each sporocarp contains eight carpospores. They are formed from the vegetative cell by a cruciate division, followed by a division parallel to the surface of the frond. These divisions give rise to two tiers of four carpospores each (Pl. XX, figs. 2*a*–2*b*; Pl. XXII, fig. 24). Occasionally the vegetative division of the sporocarp mother-cell goes one step farther; so that we find but two carpospores in each sporocarp.

Antheridia.—The antheridium-mother-cell, by a cruciate division perpendicular to the surface of the frond, gives rise to four antheridia. Each antheridium now undergoes a division parallel to the surface of the frond, then a cruciate division perpendicular to the surface of the frond, followed by another parallel division in all segments. The antheridium is now divided into sixteen parts, each of which, by a cruciate division, gives rise to four antherozoids; so that the whole antheridium now consists of sixty-four antherozoids arranged in four tiers of sixteen each (Pl. XX, figs. 3*a* and 3*b*; Pl. XXII, fig. 27). The direction of the walls laid down by the last cruciate division is often decidedly oblique, so that, when the walls separating the individual antherozoids dissolve, the groups of antherozoids are arranged in the form of a hollow sphere. The wall laid down by the first, parallel, reproductive division of the antheridium is thicker than those laid down afterwards, and only dissolves when the frond is fully ripe; so that for a long time two separate spherical groups of antherozoids exist in each antheridium.

This spherical arrangement of the antherozoids seems to be peculiar to the plants collected on the Californian shores, since specimens of *P. leucosticta* collected on the coasts of France and of Helgoland fail to show this.

Porphyra leucosticta was founded by Thuret (1864), and has since been more fully described by Janczewski (1873).

According to de Toni (1897), it is identical with *Ulva atro-purpurea* Olivi.[1]

Habitat.—On rocks and epiphytic on algæ (*Gracilaria confervoides*); lower litoral and sublitoral zones; March to May.

Distribution.—The author is aware of but a single region of the Pacific Coast where this species occurs; i. e., Monterey Bay, California (36° 45′ N. lat.).

Localities.—Pacific Grove, California (M. A. Howe!, Mrs. J. M. Weeks!, W. A. Setchell No. 5161!); Santa Cruz, California (Mrs. J. M. Weeks!, de A. Saunders!).

4. Porphyra perforata *J. Ag.*

PLATE XX, FIGS. 4*a*-10.

Till Alg. Syst. Afd. 3, VI, 1882, Ulvaceæ, p. 69.

Porphyra perforata J. AGARDH, Till Alg. Syst., Afd. 3, VI, 1882, Ulvaceæ, p. 69. HUS, in Phyc. Bor.-Amer., Fascl. XIV, COLLINS, HOLDEN & SETCHELL, No. 682, 1900. HUS, Zoe, Vol. V, 1900, p. 63.
Porphyra vulgaris ANDERSON, Zoe, Vol. II, 1891, p. 221. HOWE, Erythea, Vol. I, 1893, p. 67.
Wildemania perforata DE TONI, Syll. Alg., Vol. IV, 1897, p. 21.

Membranous, 7–70 cm. long, 3–20 cm. broad; linear-lanceolate, with undulate margin, often much expanded and laciniate; base cordate to umbilicate; attached by a disc; gray to brown-purple, becoming blue-purple on drying; monostromatic, vegetative part of frond 45–140μ thick; cells once to two and a half times as high as broad; surface jelly thick, often forming two-fifths of the thickness of the frond; monœcious, sporocarps and antheridia in irregular patches, the latter radiating towards the margin; vegetative cells often mixed in with the sporocarps, never with the antheridia; each sporocarp containing 32 carpospores, each antheridium containing 128 antherozoids.

This species is known to the author from specimens collected by Mrs. Snyder at San Diego, some of which were communicated to the late Professor J. G. Agardh, and referred by him to *P. perforata*.

It is an annual, found throughout the year, the young fronds making their appearance before the old ones have altogether disappeared. Where the fronds are exposed to

[1] Saggi Accad. di Padova III, 1, (præs. die 18 Martii 1791), p .153, Tab. I-III (fide de Toni).

the violence of the waves they disappear in the fall, something to which the dry north winds which frequently occur in California during the latter part of the year contribute not a little. The result of this is that in December and January the fronds are found in sheltered places only.

P. perforata occurs throughout the litoral zone and in the upper part of the sublitoral zone. There is some difference in form between the plants of the upper part of the litoral zone and those of the sublitoral zone. While the former, which the writer considers typical, are lanceolate, with a cordate base and a frond which is but slightly perforate, the latter are far more irregular, being much lobed and laciniate, with an umbilicate base, and are much perforate. They also seem of finer texture and are as a rule much thinner. This, however, is not always the case. The author has collected specimens in the sublitoral zone which in the sporocarpic portion of the frond measured 110μ, a thickness which corresponds to the average measurement of the sporocarpic parts of the fronds of the litoral zone. The difference in thickness seems to lie chiefly in the jelly, the latter, in the fronds of the litoral zone being as a rule more abundant than in the fronds of the sublitoral zone.

The specimens of *P. perforata* found in the sublitoral zone also possess a color different from that of the fronds of the litoral zone. While the latter are a gray- to brown-violet, the former are altogether devoid of a violet shade and appear a gray- or yellow-brown. This difference in color is very striking, and at first led to many conjectures. But in the light of the fact that herbarium specimens after having been preserved for six months or more begin to take a violet hue, a color, the various shades of which are often characteristic of the various species to which the genus owes its name, and chiefly its inclusion among the Rhodophyceæ, it seems but reasonable to suppose that the deeper violet color of the specimens of *Porphyra perforata* found in the litoral zone is due to the action of the air. What the other causes for the difference may be has not yet been solved. Perhaps the slug, which in all probability causes

the perforations, is more numerous in the sublitoral zone than in the litoral zone,[1] or perhaps the light has something to do with it; but whatever the cause, both the anatomical and morphological differences are too slight to allow us to consider the sublitoral zone form a variety of the typical *Porphyra perforata* J. Agardh.

The shape of the frond of *Porphyra perforata* is as a rule linear-lanceolate, with an undulate margin and a cordate base. But frequently more or less expanded, broadly laciniate fronds are met with, which possess an umbilicate base. Between these two extreme forms which the frond may assume, numerous transition stages are found.

The size of the frond is variable. Fruit has been found on specimens but seven centimeters long. A well-developed specimen usually measures from 50–70 centimeters. Both size and shape of the frond depend apparently largely upon local conditions. For instance, those plants which grow on the rocks in the upper part of the litoral zone seem but seldom to attain a great development, and are usually lanceolate. Those which grow in the elitoral zone or in places where they are exposed to the tow of the tide and are always, or for the greater part of the time, stretched out by the water—such as those which grow on lagoons and on long, low reefs,—are more linear-lanceolate, while, as has been said before, those growing on rocks exposed to the irregular wash of the waves are much laciniate and rather broadly expanded.

The plants are attached by a disc, the structure of which is of much interest. It agrees in all respects with that of *P. laciniata* as described by Bornet (1878). *P. perforata*

[1] Von Martens (1866), in a discussion on the use of seaweed as food by marine animals, describes some specimens of *P. vulgaris* Ag. (?) which were covered by a large number of sea-slugs (*Nasa corniculum* Olivi), and which were much perforate. In their immediate vicinity grew plants of *Grateloupia filicina* and of *Chætomorpha linum*, which were not inhabited by the slug and had not been damaged. From this he concludes that the perforate condition of the specimens of *P. vulgaris* was due to the action of the slug, which used the plants for nourishment. The slug which infests the plants *Porphyra perforata* on the Californian shores is a species of *Lacuna*. On this subject Agardh (1882) says: " * * * ; præterea frequenter foraminibus minutis rotundatis, linæm aut paucas diametro æquantibus, præcipai in disco perforatam vidi, et hoc quidem aliquando jam in speciminibus minoribus et angustis."

possesses a pronounced aureole around the point of attachment, often one centimeter in diameter.

The writer has had an opportunity to examine a large number of specimens of *P. perforata* collected at numerous localities on the shores of the Pacific Ocean, but has up to this time failed to encounter a single distromatic specimen. A thorough examination of the fronds left no doubt as to the purely monostromatic nature of the species, all consisting in the purely vegetative part of but a single layer of cells.

The thickness of the frond varies from 40 to 140μ, the difference in thickness being oftener due to a greater amount of jelly than to a difference in the height of the cells. The cells in the lowest part of the frond are usually square or nearly so, but more towards the tip they are from one and one-half to two and one-half times as high as broad.

Porphyra perforata is monœcious. Patches of sporocarps and antheridia occur here side by side, usually alternating; but not a single instance has been met with, where the antheridial and sporocarpic cells were intermixed, such as we meet with in *P. amplissima*, or where one-half of the cell formed antherozoids and the other half carpospores, such as described by Janczewski (1873) for *P. leucosticta*. The frond is often nearly entirely sporocarpic or antheridial. Vegetative cells are sometimes encountered among the sporocarps but never among the antheridia. There are also cases where one or several of the divisions of the cell in the formation of the sporocarp have not developed, so that but one or more have formed spores, while the others are to all appearances dead. The walls of such a cell are thickened, the contents are hyaline and of a deep yellow color, which sharply contrasts with the dark violet tinge of the carpospores, and the unchanged, granular contents of the vegetative cells or of the monospores. Some instances have been noted where the sporocarps occurred in long narrow patches, instead of in the broader patches usually found.

The patches of sporocarps are easily distinguished from the patches of antheridia. The former are of a uniform, dark violet color, which gradually, towards the center of the frond, fades into the gray- or brown-violet of the vegetative part of the frond. The patches of antheridia, on the other hand, are nearly colorless when fully ripe. They are usually very narrowly triangular and radiate from the center of the frond. These patches contain the unripe antheridia and are, towards the center of the frond, of a light violet color, lighter than that of the surrounding sporocarps, but deeper violet than that of the vegetative cells.

Sporocarps.—The first division of the sporocarp is cruciate and perpendicular to the surface of the frond. This is followed by a division parallel to the surface of the frond, thus giving rise to eight segments in two tiers of four each, every one of which undergoes a cruciate division, thus giving rise to thirty-two spores in two tiers of sixteen each (Pl. XX, figs. 5*a* and 5*b*; Pl. XXII, fig. 25). Rarely there is an additional parallel division in some or all the segments formed by the last cruciate division.

Antheridia.—The antheridium-mother-cell undergoes a cruciate division perpendicular to the surface of the frond, giving rise to four antheridia. The first reproductive division of the antheridium is parallel to the surface of the frond. This is followed by a cruciate division, after which another parallel division takes place; so that each antheridium now contains sixteen segments arranged in four tiers of four each. Each of these segments now undergoes first a cruciate division and then a parallel division; so that each antheridium now contains 128 antherozoids, arranged in eight tiers of sixteen antherozoids each (Pl. XX, fig. 6; Pl. XXII, fig. 28). In this species the last division is nearly always regular and fully carried out.

Sometimes the vegetative division of the antheridium-mother-cell does not take place; so that four times the number of antherozoids are formed in an antheridium.

Economic Use.—*Porphyra perforata* is one of the edible seaweeds, and is largely collected for food by Indians and Chinese on the Pacific Coast.

Habitat.—On rocks, wood and barnacles. Epiphytic on *Zostera*, *Phyllospadix*, *Nitophyllum*, *Fucus* and *Gigartina*. Throughout the litoral and in the upper part of the sublitoral zones. Found throughout the year, but during the winter months in sheltered places only.

Distribution.—At present known from the Pacific Coast only, from Alaska to Southern California (58° 30′–32° 20′ N. lat.).

Localities.—Glacier Bay, Alaska (de A. Saunders, No. 100!); Baranoff Island, Alaska (de A. Saunders, No. 130!); Shumagin Island, Alaska (de A. Saunders, No. 394!); Whidby Island, San Juan County, Washington (N. L. Gardner, No. 295!); Chehalis Bay, Washington (Ralph Emerson, No. 1792!); Crescent City, Del Norte County, California (H. Hus!); Trinidad, Humboldt County, California (H. Hus!); Duxbury Reef, Marin County, California (W. A. Setchell, No. 1055!, H. Hus, No. 81!); Farallon Islands, California (H. Hus!); Lands' End, San Francisco, California (W. A. Setchell, No. 2034!, No. 2068!, H. Hus, No. 26!, No. 69!, *et al.*); Santa Cruz, California (C. L. Anderson!); Monterey, California (W. A. Setchell, No. 5159!, H. Hus!); San Simeon Bay, California (Dr. Palmer!); Santa Barbara, California (Mrs. Cooper!); San Diego, California (Miss Reed, No. 25!); Coronado, California (Mrs. M. S. Snyder!).

5. Porphyra perforata f. segregata *Setchell & Hus.*

Zoe, Vol. V, 1900, p. 64.

Porphyra perforata f. *segregata* SETCHELL & HUS, Zoe, Vol. V, 1900, p. 64. SNYDER, in Phyc. Bor.-Amer., Fascl. XIV, COLLINS, HOLDEN & SETCHELL, No. 684, 1900.

Porphyra leucosticta TILDEN (not THURET), Amer. Algæ, Cent. III, No. 228, 1898.

This plant agrees in many respects with the species, but there are some fundamental differences, which, though they do not entitle it to a separate specific name, make it desirable to distinguish it as a distinct variety. As a rule the plant is much smaller and has a rather more umbilicate base

than *P. perforata*. The frond is much thinner, never exceeding sixty microns. The vegetative cells are smaller than those of the species, and the jelly walls separating the different cells are thicker, giving the vegetative portion of the frond when seen in surface view a most characteristic appearance.

The difference between the reproductive cells of *P. perforata* and this variety can be best seen in cross-section. It is especially noticeable in case of the antheridia. The wall laid down by the first reproductive division of the antheridium is very thick and widely separates the antherozoids into an upper and a lower group.

Distribution.—From Washington southward to Lower California (47° 30′–27° 8½′ N. lat.).

Localities.—Shillshole Bay, Seattle, Washington (Miss J. E. Tilden!); San Pedro, California (de A. Saunders, No. 1034!); San Diego, California (Mrs. M. S. Snyder!); San Roque, Mexico (G. Eisen!).

6. Porphyra perforata f. lanceolata *Setchell & Hus.*

Zoe, Vol. V, 1900, p. 65.

Porphyra perforata f. *lanceolata* SETCHELL & HUS, Zoe, Vol. V, 1900, p. 65; in Phyc. Bor.-Amer., Fascl. XIV, COLLINS, HOLDEN & SETCHELL, No. 683, 1900.

Porphyra laciniata TILDEN, Amer. Algæ, Cent. III, No. 229, 1898.

Membranous; 10–325 cm. long, 1–10 cm. broad; linear with undulate margin; base cuneate to cordate, attached by a disc; steel-gray to gray- or yellow-brown, becoming purple on drying; monostromatic, vegetative part of frond 75–150μ thick; cells one and one-half to four times as high as broad; jelly very thick, forming two-fifths to one-half the thickness of the frond; dicœcious; sporocarps containing 32 carpospores, each antheridium containing 128 antherozoids.

This variety of *P. perforata* agrees in many respects with the species. The chief difference lies in the fact that it is dicœcious. Besides this there are numerous minor differences.

P. perforata f. *lanceolata* can be distinguished at first sight from the species by its form. This is either distinctly

linear with undulate margin, or deeply laciniate, producing a two-forked frond. Sometimes a specimen, and this is true for those of even 50 centimeters long, measures but one centimeter across; but the larger specimens may be as wide as 10 centimeters. The average width is 5 centimeters, with a length of 50 centimeters. Mr. R. E. Gibbs, however, collected a specimen of this variety at the Presidio which measured 325 centimeters in length, and with a width of 10 centimeters in the broadest part. The plant was growing on a rock buried in the sand of a gently sloping beach, where it was carried back and forth by the waves, which perhaps brought about its extreme development. Something similar is met with in the linear-lanceolate fronds of *P. perforata* when growing in lagoons.

The base is cuneate to cordate, and is but rarely umbilicate. The plant is attached by a disc, the structure of which is identical with that of the disc of *P. perforata*.

The color of the fronds varies considerably. As a rule the fronds are steel-gray to gray- or yellow-brown; sometimes they are in part a bright green. In mature fronds the edges, if the frond is antheridial, are yellow and appear much swollen. The latter is caused by the swelling of the jelly preparatory to breaking down and setting the antherozoids free. The sporocarpic frond is usually of a darker color, the edges having a reddish brown appearance.

The difference between antheridial and sporocarpic fronds is brought out still better on drying, when the antheridial fronds become distinctly yellow at the edges, and the sporocarpic fronds, in the region of the sporocarps, red-violet. The latter fronds are also less shiny than the antheridial fronds.

Occasionally forked fronds are met with; and in such cases it is not unusual to find one fork bearing antheridia, while the other is strictly sporocarpic. These subdiœcious fronds form a connecting link between the form and the species proper.

The number of divisions in the sporocarps and antheridia seems to be the same as in *P. perforata*.

Habitat.—*P. perforata* f. *lanceolata* is an annual, occurring throughout the year, but in winter found only in sheltered places. It usually grows on rocks in the highest part of the litoral zone. In some cases it has been found on but a single rock for a distance of over a mile. This was in December, but the rocks in the same locality (Fort Point, Presidio, San Francisco) were found to be thickly covered with it in June of the next year.

Distribution.—Up to now this plant has, to the author's knowledge, been found in but two localities, both in California (37° 47′–36° 45′ N. lat.).

Localities.—Lands' End, San Francisco, California (W. A. Setchell!, H. Hus!, R. E. Gibbs, Miss J. E. Tilden!); Carmel Bay, Monterey County, California (W. A. Setchell!, R. E. Gibbs!).

7. Porphyra nereocystis *Anderson.*

PLATE XX, FIGS. 11*a*-12.

Zoe, Vol. III, 1892, p. 148.

Porphyra nereocystis ANDERSON, Zoe, Vol. II, 1891, p. 221, (name only); Vol. III, 1892, p. 149 (descr.). HOWE, Erythea, Vol. I, 1893, p. 67. SETCHELL, in HOLDEN, COLLINS & SETCHELL, Phyc. Bor.-Amer., Fascl. XII, No. 583, 1899. HUS, Zoe, Vol. V, 1900, p. 65.

Pyropia californica J. G. AGARDH, Anal. Algol., Cont. V, 1899, p. 153.

Fronds 3–270 cm. long, 2–40 cm. broad, linear to oblong, with laciniate margin and obtuse or cucullate base; attached by a disc; fronds red to purple, monostromatic, vegetative part 25–60*μ* thick, little jelly between the cells; fronds monoecious, antheridia forming light-colored, sharply defined spots and streaks among the dark-colored sporocarps, 32 carpospores in each sporocarp, 128 antherozoids in each antheridium.

This species, first mentioned by Dr. Anderson of Santa Cruz, California, is one of the largest of known species of *Porphyra*. It attains a great length, specimens 90 centimeters long being by no means rare. From notes appended to a fragment of one specimen it was gathered that the whole specimen measured 270 centimeters. The shape is fairly constant. The younger specimens are lanceolate and have an even margin. Older specimens are oval and

have a cucullate base. The aureole around the disc is very pronounced in this species. The frond is not plicate, but on drying several folds appear in some of the older specimens, owing to the concave nature of the frond caused by the cucullate base. The margin in the older fronds is much laciniate.

The fronds are monostromatic in the vegetative part. No deviation from this has been observed. The cells are square with rounded angles, or may be twice as high as broad or twice as broad as high.

Sporocarps.—The mature frond becomes gradually changed into reproductive cells of which the sporocarps form the majority. They can be readily recognized by their deep red color, and are sharply outlined against the lighter colored antheridia. The sporocarp first divides cruciately, giving rise to four cells. Each of these now undergoes a parallel division, followed by a cruciate division; so that in each sporocarp we finally have thirty-two carpospores arranged in two tiers of sixteen each (Pl. XX, fig. 11*c*; Pl. XXII, fig. 25).

Antheridia.—They are found in larger or smaller, irregular, light-colored patches among the sporocarps. The antheridium-mother-cell, by a cruciate division, gives rise to four antheridia, each of which by alternating parallel and cruciate divisions, as in *P. perforata*, give rise to 128 antherozoids, arranged in eight tiers of sixteen each.

Economic Use.—This plant is largely collected by Chinamen on the Californian coast, along with *P. perforata*. From what could be learned, it seemed that it is much more esteemed than the latter.

Habitat.—This species is found attached to the stipes of *Nereocystis lütkeana* in from three to five fathoms of water, and is also found on rocks. After a storm it is frequently found floating.

Distribution.—*Porphyra nereocystis* has a very wide range, having been collected at St. Paul, Kadiak Island, Alaska, and as far south as San Pedro, California (57° 30′–33° 40′ N. lat.).

Localities.—St. Paul, Kadiak Island, Alaska (W. A. Setchell and A. A. Lawson, No. 5149!); Coupeville, Washington (N. L. Gardner!); Bolinas Bay, Marin County, California (W. A. Setchell, No. 1275!); Monterey Bay, California (W. A. Setchell, No. 3065*a*!; C. L. Anderson!); San Pedro, California (de A. Saunders!).

8. Porphyra naiadum *Anderson.*

PLATE XXI, FIGS. 19-22.

Zoe, Vol. III, 1892, p. 148.

Porphyra naiadum ANDERSON, Zoe, Vol. III, 1892, p. 148. HOWE, Erythea, Vol. I, 1893, p. 67. MCCLATCHIE, Proc. So. Cal. Acad., Vol. I, 1897, p. 356. TILDEN, Amer. Algæ, Cent. III, No. 231, 1898. SETCHELL in Phyc. Bor.-Amer., Fascl. XIII, COLLINS, HOLDEN & SETCHELL, No. 632, 1899. HUS, Zoe, Vol. V, 1900, p. 66.

Porphyra coccinea AGARDH, J. G., Till. Alg. Syst., Afd. 3, VI, 1882, p. 58; in part (as to the Californian specimens ?).

Porphyra sp. RUPRECHT, Neue oder unbek. Pflan. d. N. Th. d. St. Oceans, 1852, p. 65.

Fronds 2-10 cm. long, obovate when young, oblanceolate when older; base cushion-shaped; fronds wine-red to blue-purple; monostromatic, vegetative part 25-30μ thick, cells square or slightly higher than broad, 15-20μ high; surface jelly measuring about 5μ, little jelly between the cells; fronds dicœcious ?; sporocarps with eight carpospores.

The first mention of this species of *Porphyra* was probably made by Ruprecht (1852), who speaks of a parasitic *Porphyra* occurring on *Phyllospadix scouleri.* It was first recognized as a distinct species by Dr. Anderson in 1892; but for reasons given below it is evident that it was known before that time to J. G. Agardh. Formerly it had been distributed along with *P. nereocystis* and *P. perforata* as *P. vulgaris*, but, as Dr. Anderson says, "without much more reason than our early botanists had for placing all seaweed in the genus *Fucus.*" It is found growing on *Phyllospadix* at extreme low-water mark, and in sheltered and exposed places alike. The plants occurring on a single blade of eel-grass are usually so numerous that the color of the adult fronds literally hides the green color of the eel-grass. It is interesting to note that most of the fronds

occurring on *Zostera* usually attach themselves to the edges of the blade, forming a broad fringe.

In the spring of 1898, Dr. Setchell called the attention of the author to some plants of *Phyllospadix*, the leaves of which were densely covered by numerous small, reddish-brown, cushion-shaped growths, which he suspected to stand in some relation to *P. naiadum*. On examination, these cushions proved to consist of a number of angular cells (Pl. XXI, fig. 19), the outer layers of which contained chromatophores, which were deeply lobed, somewhat after a stellate fashion, resembling those described by Schmitz (1882) for *Helminthocladia purpurea*. Each cell of the layer adjacent to the substratum possessed a short rhizoid, which apparently attached itself to the cuticle of the host-plant. In the beginning of February, these cushions begin to show short hair-like projections. These projections are composed of cells placed in a single narrow layer. The youngest of them consist of but a few cells placed in a single row. Older specimens show that these cells divide in two directions, giving rise to a monostromatic frond which proves to be *P. naiadum* (Pl. XXI, fig. 22).

The cells of the lower part of the frond, unlike those at the base of fronds of other species of *Porphyra*, do not produce hypha-like projections. Even in the oldest plants no sign of this was to be found. This prothalloid base is different from the bases of all known species of *Porphyra*, as described by Berthold, Thuret, and others, with the exception, perhaps, of one described by J. G. Agardh (1882) under the name *P. coccinea*.

The size of the frond varies somewhat. The young fronds are obovate, measuring about one centimeter across; but the frond gradually broadens and lengthens, finally assuming an oblanceolate shape, being from four to ten centimeters long and two or three centimeters broad. There is a considerable difference in size and color between specimens growing in sheltered places and those growing in the open. In fact, we may distinguish between two forms—*a*, *minor* and *b*, *major*. Under the first, we understand the

form of *P. naiadum* found on *Phyllospadix*, growing in exposed places, with a smaller, rather orbicular frond and a laciniate margin; while by *P. naiadum* f. *major*, we mean the plant which grows on *Zostera*, in lagoons and other sheltered places, with a larger, broader frond, and which usually possesses an even margin.

Only one form of fruit was found, viz.—sporocarps. As in the case of the reproductive bodies of the other species of *Porphyra*, they are here first formed in the region of the tip, gradually spreading over the whole frond.

Sporocarps.—In the formation of the sporocarp, the first division is cruciate, followed by a division parallel to the surface of the frond, thus giving rise to eight spores in two tiers of four each (Pl. XXI, figs. 21*a* and 21*b*; Pl. XXII, fig. 24). At the time of the ripening of the sporocarp the jelly around the spores swells, thus making the frond a few microns thicker, but finally dissolves as does the outer jelly-wall, setting the spores free.

Dr. Anderson suggests that small molluscs, in rasping for food on eel-grass, cause abrasions, "in which the spores of this *Porphyra* find a place to adhere;" but careful sectioning has failed to show any injury to the epidermal layer of the eel-grass at the places of attachment of very young fronds.

Habitat.—The plant has been collected throughout the year. It grows exclusively on *Phyllospadix* and *Zostera*. In sheltered places we find it all through the winter, but where the eel-grass is exposed to the direct influence of the tides, the *Porphyra* disappears about August, to reappear in the prothalloid form about the latter part of January.

Distribution.—This plant appears to be limited to the Pacific Coast, occurring from Washington to Southern California (48° 10′–32° 20′ N. lat.).

Localities.—Puget Sound (de A. Saunders!); Coupeville, Washington (N. L. Gardner!); Ballard Beach, Seattle, Washington (Miss J. E. Tilden!); Crescent City, Del Norte County, California (H. Hus!); Fort Ross, Sonoma County, California (W. A. Setchell, No. 1789!); Duxbury

Reef, Marin County, California (W. A. Setchell, No. 1037!, No. 1276!, H. Hus, No. 80!); Farallon Islands, California (T. W. Blankinship); Land's End, San Francisco, California (W. A. Setchell, No. 1118!, H. Hus!); Monterey, California (W. A. Setchell, No. 5158!, C. P. Nott!, Mrs. Bush!, R. E. Gibbs!, H. Hus!); Santa Cruz, California (C. L. Anderson!); San Pedro, California (A. J. McClatchie!); San Diego, California (Miss Reed, No. 107!).

9. Porphyra amplissima *(Kjellman) Setchell & Hus.*

PLATE XX, FIGS. 13*a*–13*b*.

Zoe, Vol. V, 1900, p. 67.

Porphyra amplissima (KJELLMAN) SETCHELL & HUS. HUS, Zoe, Vol. V, 1900, p. 67. GARDNER, in Phyc. Bor.-Amer., COLLINS, HOLDEN & SETCHELL, Fascl. B, No. XLIX, 1901.

Diploderma amplissimum KJELLM., The Algæ of the Arctic Sea, p. 188, Pl. 17, figs. 1–3; Pl. 18, figs. 1–8, 1883.

Diploderma amplissimum f. *typica* FOSLIE, Cont. to Knowledge of the Marine Algæ of Norway, I, 1890, p. 56.

Wildemania amplissima FOSLIE, Cont. to Knowledge of the Marine Algæ of Norway, II, 1891, p. 14. DE TONI, Sylloge Algarum, Vol. IV, 1897, p. 24.

Fronds membranous, 20–60 cm. long, 10–15 cm. broad, broadly elliptical to ovate-lanceolate, with much undulate margins deeply folded, the folds usually reaching to the median line of the frond; color deep red-purple; base slightly cuneate, sometimes cordate, sessile with small disc; fronds distromatic, 50–80μ thick; cells in cross-section square or slightly higher than broad, with rounded angles; surface jelly 5–10μ thick; fronds monœcious, antheridia and sporocarps intermixed in a marginal zone, sometimes together with vegetative cells; each sporocarp consisting of 4 or 8 carpospores, each antheridium consisting of 16 antherozoids.

This alga is the *Diploderma amplissimum* of Kjellman (1883), agreeing in every respect with both drawings and description. It varies considerably in size. Kjellman suggests that the greater size is due to the floating condition. It is, however, doubtful if plants of this species continue to grow after being torn away. The author, not having collected this species, is unable to express an opinion, but judging from the fact that plants of other species of

Porphyra when found floating are in a very poor condition it seems very unlikely. One would rather be inclined to ascribe the greater development of specimens of *P. amplissima* to little exposure, since, judging from notes appended to specimens examined, it appears to be due to the plants growing in undisturbed water, while specimens collected in exposed places always were small.

The shape of the fronds is to a certain extent variable. The extreme forms are broadly elliptical and ovate-lanceolate. Between these two a large number of intermediate forms may be noted. The diversity in form is perhaps due to a great extent to mechanical influences. An important factor is also the age of the specimen. The older, longer plants seem to possess a greater lateral development, out of proportion to the increase in length.

The color seems likewise to depend on the age of the plant. The younger specimens vary from lake to deep red-purple, but the color seems to fade out in the older specimens, which are often a faint brownish red.

The fronds are densely folded, the folds extending to the median line of the frond. The margin is smooth or laciniate, a matter which also probably depends on greater or less exposure.

The base is as a rule more or less cuneate. In some specimens the base is strongly cordate. Many intermediate forms may be met with. Our plants from Alaska are not stipitate and in this regard differ from Kjellman's description. They are sessile upon a small disc, much smaller than the one we find in *Porphyra perforata*.

The thickness of the frond varies from fifty to eighty microns. The majority of the specimens, however, measured about sixty microns, both in the vegetative and in the reproductive parts. The shape of the cells in the middle of the frond is, in cross-section, square or slightly higher than broad.

The plant is as a rule monœcious, sporocarps and antheridia occurring side by side, as shown in figs. 13*a* and 13*b*. Frequently one cell develops into antheridia, while the

corresponding cell on the other side of the frond becomes a sporocarp. Sometimes vegetative cells are found among the reproductive cells to a greater or less extent. Sometimes specimens are met with which are diœcious or nearly so; but the majority of the fronds are monœcious.

Sporocarps.—The number of spores in each sporocarp is four or eight. The sporocarp divides into four parts by a cruciate division. The division may take place in one direction only (perpendicular to the surface of the frond), or one line only may be perpendicular and the other parallel to the surface. These divisions would give rise to four spores in one tier or to four spores in two tiers of two spores each. A cruciate division perpendicular to the surface seems to occur most frequently. In this case a third division, parallel to the surface of the frond, sometimes takes place, thus giving rise to eight spores in two tiers of four each (Pl. XX, figs. 13*a* and 13*b*; Pl. XXII, fig. 24). In making these observations care was always taken to select an absolutely ripe portion of the frond.

Antheridia.—The division of the antheridium-mother-cell goes one step further than that of the sporocarp-mother-cell, a cruciate vegetative division perpendicular to the surface of the frond taking place before the formation of the antheridium proper is begun. A sporocarp therefore corresponds to four antheridia. Each of these antheridia surrounds itself with a wall of jelly. By subsequent divisions, the first parallel to the surface of the frond, the second a perpendicular cruciate division, and the third again parallel, there arise sixteen antherozoids arranged in four tiers of four each (Pl. XX, figs. 13*a* and 13*b*; Pl. XXII, fig. 26). This number may, however, vary, and from two causes. The division of the antheridium-mother-cell is sometimes incomplete, so that each antheridium becomes four times as large as the normal ones. Usually in such cases an additional reproductive division takes place, so that each antheridium possesses sixty-four antherozoids. Frequently also the divisions in the antheridium are incomplete, so that the number of antherozoids is less than sixteen.

Habitat.—On rocks in the sublitoral zone. Also found floating. June, July, August.

Distribution.—From Alaska southward to Washington (60°–48° 10′ N. lat.).

Localities.—Orca, Alaska (W. A. Setchell and A. A. Lawson, No. 5165!, de A. Saunders, No. 259*a*!); Unga, Alaska (W. A. Setchell and A. A. Lawson, No. 5047!); Amaknak Island, Alaska (W. A. Setchell, No. 3268!); Coupeville, Washington (N. L. Gardner, No. 47! No. 280! No. 199*b*!).

10. Porphyra miniata *(Lyngb.) Ag.*

Porphyra miniata (LYNGB.) AGARDH, C., Syst. Algarum, 1824, p. 191. AGARDH, J., Till Alg. Syst., VI, 1882, p. 59.
Ulva miniata LYNGBYE, Hydrophyt. Dan., 1819, p. 29, Tab. 6, D.
Diploderma miniatum KJELLM., Algæ of the Arctic Sea, 1883, p. 189.
Wildemania miniata FOSLIE, Cont. to Knowledge of Marine Algæ of Norway, II, 1891, p. 14 (in part); DE TONI, Sylloge Algarum, Vol. IV, 1897, p. 23.

11. Porphyra miniata f. cuneiformis *Setchell & Hus.*

PLATE I, FIG. 14.

Porphyra miniata f. *cuneiformis* SETCHELL & HUS. HUS, Zoe, Vol. V, 1900, p. 68.

Fronds membranous, 15–50 cm. long, 4–15 cm. broad, lanceolate with undulate, often crenulate, margins; color red-purple; base strongly cuneate, attached by a disc; fronds distromatic, often monostromatic near edge in vegetative part, 30-75μ thick; cells square to twice as long as broad; surface jelly 10–12.5μ thick; fronds monœcious, antheridia and sporocarps intermixed in a marginal zone, gradually spreading over the whole frond, each sporocarp containing 4 carpospores, each antheridium consisting of 8 antherozoids.

Nearly two hundred specimens of this species, collected at different times on the Pacific Coast, agreed essentially in all respects among themselves as well as with specimens of *Porphyra miniata* from the Atlantic coast; among others with those distributed in the Phyc. Bor.-Amer. by Mr. Isaac Holden (No. 377). Agardh's notes on *Ulva purpurea (P. miniata)* are too meager to give a definite clue in the determination of the species, especially since the number

of antherozoids in each antheridium, which the author proposes as a criterion for the species of *Porphyra*, is not mentioned. But, since the specimens do not agree with the description of *Diploderma amplissimum* by Kjellman, and since it is evident that they cannot belong to *P. tenuissima* (Strömf.) S. & H. or *P. abyssicola* Kjellm., the writer has deemed it advisable to assign the name *P. miniata* to these specimens, until such time as others who have access to the type-specimens may determine its rightful position.

This species differs from the preceding one in color, in being less deeply folded, and in possessing a crenulate margin which gives the fronds a very characteristic appearance.

It differs from Kjellman's description of *P. miniata* in being monœcious, and in possessing a strongly cuneate base. These two characters were deemed sufficient to differentiate the plant under a form name.

Sporocarps.—The sporocarps contain four carpospores, formed by a cruciate division of the mother-cell (Pl. XX, fig. 14; Pl. XXII, fig. 23).

Antheridia.—The antherozoids appear here in groups of eight arranged in two tiers of four each, thus showing in cross-section four antherozoids (Pl. XX, fig. 14; Pl. XXII, fig. 25). At first it was thought that this appearance was due to an unripe condition of the part of the frond examined, but a large number of sections from carefully selected portions of the frond showed the same character.

Sometimes sixteen antherozoids were found in one antheridium; but in that case they were arranged in two tiers of eight each. This showed that the double number of antheridia was caused by the fact that the last vegetative division in the antheridium-mother-cell did not take place.

Habitat.—Found floating.

Distribution.—From Alaska southward to middle California (60°–36° 45′ N. lat.).

Localities.—Lowe Inlet, Gulf of Alaska (de A. Saunders, No. 20!); Coupeville, Washington (N. L. Gardner, No. 199*a*!); Monterey Bay, California (Mrs. J. M. Weeks!).

12. Porphyra tenuissima *(Strömf.) Setchell & Hus.*

Zoe, Vol. V, 1900, p. 68.

Porphyra tenuissima (STRÖMF.) SETCHELL & HUS. HUS, Zoe, Vol. V, 1900, p. 68.

Diploderma tenuissimum STRÖMF., Bot. Centralbl., Bd. XXVI, 1886, p. 173; Om Algvegetationen vid Islands Küster, 1886, p. 33.

Diploderma amplissimum f. *tenuissima* FOSLIE, Contribution to Knowledge of the Marine Algæ of Norway, I, 1890, p. 56.

Porphyra miniata f. *tenuissima* ROSENVINGE, Grønlands Havalger, 1893, p. 827; Les Algues Marines du Grœnland, 1894, p. 83.

Wildemania tenuissima DE TONI, Sylloge Algarum, Vol. IV, 1897, p. 23.

Fronds membranous, 25 cm. long, 14 cm. broad, round-ovate, not lobed, with undulate margin, folded; base cordate, sessile, with small disc; color delicate pink; fronds distromatic, 25μ thick, except at the base, where they measure 65–75μ; cells near base square to twice as broad as high, 10–15μ broad, cells near edge of frond three to four times as broad as high, 20μ broad, with rounded angles; surface jelly 2.5μ, little jelly between the cells; fronds monœcious, antheridia and sporocarps intermixed in a marginal zone, 4 carpospores to each sporocarp, 8 antherozoids to each antheridium.

The description is drawn from several specimens collected by Mr. de Alton Saunders when with the Harriman Alaskan expedition during the summer of 1899. The study of a large number of specimens of other species of *Porphyra* has led to the conviction that, to describe a species from but a few specimens is, to say the least, a very unreliable method of procedure; and were it not for the fact that the author had access to a number of European specimens of *P. tenuissima*, as well as to Strömfelt's ample description (1886), with which the specimen in question agreed in nearly every respect, such a course would not have been undertaken. In matters such as length and breadth of the frond, the evidence submitted is of course not conclusive. It must be said that among our specimens are the largest which have as yet come to our notice. The European specimens are less than half the size. In the collection of *Porphyra* kindly placed at our disposal by Mr. Collins, a specimen was found, collected at Nahant, Massachusetts, which was originally determined as *P. leucosticta*[1] and afterwards designated *P. miniata*,[2] but which is evidently *P. tenuissima*.

[1] Bull. Torrey Bot. Club, Vol. IX, 1882, p. 70.
[2] Bull. Torrey Bot. Club, Vol. XI, 1884, p. 131.

It is about fourteen centimeters long and ten centimeters broad. It is the writer's opinion that the specimen in question belongs to the set distributed in the Phycotheca Generalis, Fascl. I, No. 8, and referred to by Foslie (1890) when speaking of a specimen of *P. miniata* communicated to him by Collins, and collected at Nahant, Massachusetts.

The shape of the fronds of *P. tenuissima* seems eminently constant. Most specimens are roundish ovate, and are not lobed in the slightest. The fronds are folded, more so than in *P. miniata* but less than in *P. amplissima*. The color is a delicate pink. This character together with the thinness of the fronds affords the means of readily recognizing *P. tenuissima* from other species of *Porphyra* having the *miniata*-type of antheridia.

The base seems to be cordate, and is attached by a small disc.

The fronds are distromatic throughout. There is some variation in the thickness of the frond and in the shape and size of the cells. While toward the edges the frond is but little more than twenty to thirty microns thick, and vegetative cells, where they occur, are from three to four times as broad as high, the cells being fifteen to twenty microns high and from four to six microns broad, in the lower part of the frond, near the base, the thickness varies from sixty-five to seventy-five microns, and the cells are sometimes square, sometimes twice as broad as high, being about twenty microns broad and from ten to twenty microns high. In this regard our specimens from the west coast agree fully with the description of Strömfelt.

But it is different with the monœcious character. Strömfelt describes this species as diœcious, only having found sporocarps. But what we know of the variability of the different species of *Porphyra* in this respect does not warrant us in referring these specimens other than to *P. tenuissima*, awaiting a more extended comparison between our forms and those of northern Europe.

The antheridia and sporocarps are intermixed in a marginal zone. Occasionally one or more vegetative cells are found; but this is rarely the case. The antheridia are present in predominating numbers. The reproductive part of the frond has very much the appearance of that of *P. miniata* in regards arrangement of sporocarps and antheridia. Often a cell on one side of the frond will form antheridia, while the corresponding cell on the other side of the frond will form carpospores.

Sporocarps.—The first division of the sporocarp is perpendicular to the surface of the frond. This is followed by a division at right angles to it and perpendicular to the surface of the frond, or nearly so, thus giving rise to four carpospores in a single tier, showing in cross-section two carpospores (Pl. XXII, fig. 23). The divisions are more or less regular. They may take place simultaneously, thus forming a cruciate division, or they may be consecutive, and the second division in one half of the carpospore is independent of that in the other half.

Antheridia.—The vegetative division in the antheridium-mother-cell goes one step farther than in the sporocarp-mother-cell, a cruciate division perpendicular to the surface taking place. After this a division parallel to the surface of the frond occurs in each antheridium, followed by a very regular cruciate division perpendicular to the surface of the frond. This last division, however, may precede the parallel division. These divisions finally give rise to eight antherozoids in each antheridium, arranged in two tiers of four each, so that the original antheridium-mother-cell now contains four groups of eight antherozoids each, each group corresponding to a carpospore (Pl. XXII, fig. 25).

From the above it will be seen that *P. tenuissima* possesses the *miniata* type of antheridia. It is for this reason that the writer doubts the wisdom of Foslie's action in making *P. tenuissima* a form of *P. amplissima*.

Habitat.—Epiphytic on algæ; also on rocks?.

Distribution.—As yet only known on the west coast of North America from Alaska (59° 40′-55° N. lat.).

Localities.—Yakutat Bay, Alaska (de A. Saunders, No. 214!); Baranoff Island, Alaska (de A. Saunders, No. 136!, No. 137!, No. 148!); Shumagin Island, Alaska (de A. Saunders, No. 384!).

13. Porphyra abyssicola *Kjellm.*

Algæ of the Arctic Sea, 1883, p. 191.

Porphyra abyssicola KJELLMAN, Algæ of the Arctic Sea, 1883, p. 191; Pl. 17, fig. 4; Pl. 18, figs. 10–11. FOSLIE, Cont. to Knowledge of the Marine Algæ of Norway, I, 1890, p. 60. HUS, Zoe, Vol. V, 1900, p. 68. DE TONI, Sylloge Algarum, Vol. IV, 1897, p. 14.

Fronds membranous, 2–10 cm. long, 1–4 cm. broad, lanceolate-obovate, with undulate margins; color livid cerise; base cuneate, sessile with small disc; distromatic, often monostromatic, 25μ thick; cells of the middle of the frond square or nearly twice as long as broad, with rounded angles; fronds monœcious, often diœcious; when monœcious antheridia and sporocarps intermixed in a marginal zone, 2–4 carpospores to each sporocarp, 8 antherozoids to each antheridium.

This species is known to the author by some twelve specimens found among a number of specimens of *P. naiadum* growing on *Zostera*. They agree in every respect with the description of *P. abyssicola* of Kjellman. Only a few of the specimens were fertile. On examination it was found that some were monœcious and others diœcious.

The specimens examined grew on *Zostera*, and were easily distinguished by their more elongated frond and by the absence of the characteristic cushion-shaped base of *P. naiadum*. The plants of *P. abyssicola* were also deeply though sparsely plicate.

In this species the difficulty of assigning it to the monostromatic or distromatic group makes itself more felt than in any other case. The specimens which were available for examination were for the greater part distromatic, and I hesitated considerably before assigning them to *P. abyssicola*. But it must be remembered that plants of this group vary considerably in this regard, and since all other characters were those of Kjellman's *P. abyssicola*, they were finally assigned to this species.

The possibility that a species might possess plants some of which were monostromatic while others were distromatic, or that one plant might be monostromatic in one part and distromatic in the other, and this distromatic nature not due to reproductive division, was evidently not considered by Kjellman when he divided the genus *Porphyra* into *Porphyra* proper and *Diploderma*. Not only do we meet monostromatic and distromatic forms in *P. abyssicola*, but *P. miniata* and *P. tenuissima* also exhibit this character in a greater or less degree. It therefore seems to the writer unwarranted to separate such closely allied species by a criterion which does not hold true in all cases; and he fully concurs in the action of Rosenvinge, who, after finding many intermediary forms, reunited the two subgenera. But the writer cannot agree with Rosenvinge in uniting *P. miniata*, *P. amplissima*, *P. tenuissima*, and *P. abyssicola* under *Porphyra miniata*, since there exist too many characteristic differences, of which the difference in the number of antheridia is but one. The advantage of having but one genus is not only exemplified in the *miniata* group, but is also shown in others. Not only are several distromatic species monostromatic during some part of their existence, but all monostromatic species become practically distromatic when in fruit. Besides, the monostromatic nature of the frond does not alter the external appearance by which we can always readily recognize a member of the genus *Porphyra*.

Sporocarps.—The formation of the sporocarps of *P. abyssicola* is apparently identical with that of the carpospores of *P. miniata*, so that, when ripe, each sporocarp contains four carpospores (Pl. XXII, fig. 23).

Antheridia.—Like the sporocarps, the antheridia of *P. abyssicola* agree entirely with those of *P. miniata*, each antheridium containing when ripe eight antherozoids (Pl. XXII, fig. 25).

Habitat.—On *Zostera*; also on rocks (fide Kjellman).

Distribution.—As yet known on the Pacific Coast from but a single locality (48° 10′ N. lat.).

Locality.—Whidby Island, San Juan County, Washington (N. L. Gardner, No. 273*a*!).

14. Porphyra variegata *Kjellm. in litt.*

PLATE XXI, FIG. 18.

Zoe, Vol. V, 1900, p. 69.

Porphyra variegata KJELLM. *in litt.*
Diploderma variegatum KJELLMAN, K. Sv. Vet. Akad. Handl., Bd. XXIII, 1889, p. 33, Pl. II, figs. 1-4.
Wildemania variegata DE TONI, Nuova Notarisia, Annata V, 1890, p. 148; Sylloge Algarum, Vol. IV, 1897, p. 23.

Fronds membranous, 10-80 cm. long, 2-20 cm. broad, obovate to lanceolate, with slightly irregular margin, slightly undulate, base cuneate to obtuse, or even cordate, sessile with small disc, areolate; color rich crimson, often variegated; fronds distromatic, 100-220μ thick; cells in immediate vicinity of attachment spherical or square with rounded angles, other vegetative cells from 3-5 times as high as broad, 30-60μ high, 3-30μ broad, with rounded angles; surface jelly thick, 20-45μ; cell-wall thick, composed of several layers of jelly; fronds dicœcious, sporocarps spreading over the whole frond, more or less intermixed with vegetative cells; each sporocarp containing from 8 to 32 carpospores.

The average length of some hundred specimens of *Porphyra variegata*, collected at various localities on the Pacific Coast is 20 to 25 centimeters. Several plants in our herbarium fall below that mark, measuring from ten to fifteen centimeters, while a few attain a length of from forty to eighty centimeters. These measurements all refer to dried specimens, and judging from the fact that plants of this species, when soaked out, increase to about twice the size of the dried specimen, it is supposed that the plants contract considerably when drying; something which has also been indicated by Kjellman (1889), who was able to collect *Porphyra variegata* himself. The author was less fortunate, and had to depend on dried specimens collected by others. The impossibility of examining freshly gathered specimens was keenly felt at first,—since specimens as soon as soaked out disintegrated so rapidly as to make the preparation of satisfactory sections out of the question. After a number of experiments, it was found that if the dried

plants were treated with a boiling concentrated solution of corrosive sublimate the cells retained their natural position and size, at least sections from material treated in this manner presented apparently a normal appearance. As a mounting medium twenty per cent. glycerine in water was found to act very satisfactorily; but this was afterwards superseded by Farrant's Medium, on account of the easier manipulation.

The shape of the younger fronds of *P. variegata* is obovate. The larger fronds are more or less lanceolate, and are but seldom lobed, though a few cases were noted in which the frond exhibited considerable lateral development, together with a much laciniate appearance. The fronds are slightly undulate and possess as a rule a cuneate to obtuse base. A few specimens exhibited a cordate base. The plants are attached by a small disc, surrounded by a slight aureole. In this regard the plants differ from the description given by Kjellman, who calls them substipitate. However, a careful examination of our specimens has failed to reveal a single instance of this.

The sterile frond has a red-purple color, but the fertile frond presents a variegated appearance, especially towards the center of the frond, while along the edges it is a rich, uniform crimson, sharply contrasting with the yellowish rim, some two to three millimeters broad, which surrounds the fully ripe frond till near the base. This yellowish rim consists of an empty network of gelatinous cell-walls, which formerly contained the carpospores. The latter, when ripe, left the frond because of the partial dissolution of the surrounding cell-walls. This network is full of infusoria.

The crimson edges consist almost entirely of sporocarps, with but few vegetative cells intermixed; while the variegated appearance of the central part of the frond is due to an intermixing of groups of deeply stained sporocarps with a greater or lesser number of colorless vegetative cells. In the sterile frond the vegetative cells are all colored, but not intensely so. It has been suggested that the lack of color in

the vegetative cells mixed with the sporocarps is due to the absorption of the coloring matter by the reproductive cells; but this hypothesis is not altogether satisfactory.

The fronds are distromatic throughout, and any one frond is fairly constant in thickness. But among the various fronds a great difference in thickness may be noted, some measuring but 100 microns while others measure as much as 220 microns. But if the thickness of each frond is constant throughout, the size and shape of the cells in the various parts of the frond are exceedingly variable. While at the base, the cells, especially those possessing the hypha-like projections which form the disc, are spherical or pear-shaped, the cells a short distance away from the place of attachment are square, measuring in each direction from thirty to fifty microns. But the vegetative cells among the fully ripe sporocarps are from thirty to sixty microns high and but from three to ten microns broad. They appear to have assumed this shape owing to much lateral pressure, being much elongated and otted dumb-bell shaped (Pl. XXI, fig. 18).

The great thickness of the surface jelly and the peculiar markings of the walls of the vegetative cells are characteristic of this species. The cell-wall appears to consist of layers of jelly of different density. These seem to denote stages in the cells existence; but what these stages are must at present be left to conjecture.

This species appears to be diœcious, no antheridia having been found.

Sporocarps.—The first division of the cell destined to become a sporocarp is cruciate, which is followed by a division parallel to the surface of the frond in each of the subdivisions of the sporocarp, thus giving rise to eight sporocarps arranged in two tiers of four each. Often another cruciate division takes place, giving rise to thirty-two carpospores (Pl. XXI, fig. 18).

In *Porphyra variegata* the jelly, especially that which forms the partitions between the individual carpospores,

seems to disintegrate more readily than in any other species of *Porphyra*, with the exception perhaps of *P. leucosticta*. Consequently we find in the arrangement of the fully ripe carpospores an irregularity such as is met with in the arrangement of the antherozoids in *P. leucosticta*. The carpospores arrange themselves along the lines of least pressure, and consequently the whole group becomes spherical and each carpospore more or less polygonal.

Habitat.—On rocks. Zone ? August.

Distribution.—As far as the author is aware, this species has been reported but once, and that by Kjellman (1889), its author, from Bering Island. Judging from the table of distribution in the paper above referred to, *P. variegata* seemed to be very local, and the impression was created that it belonged to colder waters. Much surprise was therefore experienced when it was reported from San Pedro, California. Later it was found at Coupeville, Washington (55°–33° 40′ N. lat.). It was not observed in Alaska.

Localities.—Bering Island (Kjellman); Coupeville, Washington (N. L. Gardner, No. 177!, No. 179!); Monterey Bay, California (C. L. Anderson!, C. P. Nott, No. 863!, Mrs. J. M. Weeks!); San Pedro, California (de A. Saunders, No. 1206!, No. 1207!).

15. Porphyra occidentalis *Setchell & Hus.*

PLATE XXI, FIGS. 15*a*–17*b*.

Porphyra occidentalis SETCHELL & HUS, Zoe, Vol. V, 1900, p. 69.

Fronds 15–30 cm. long, 1.5–5 cm. broad, linear, with slightly crenulate margin and a cuneate to orbicular base, sessile on small disc; color dull red; fronds distromatic, vegetative part of frond, 45–75μ thick, cells square or once and a half to twice as broad as high, 12–15μ high, 12–30μ broad, surface jelly 10–15μ thick; apparently dioecious, antheridia forming a yellowish margin, measuring 65–75μ in cross-section, 64 antherozoids in 4 tiers of 16 antherozoids in each antheridium.

This species is known to the author by but a few specimens collected by Mrs. Weeks at Carmel Bay, Monterey County, California. The plant has very much the external

appearance of a young frond of *P. nereocystis*, being linear to slightly lanceolate in the more developed fronds. The margins are slightly crenulate and more or less irregular, but the frond is not lobed or even laciniate. The base is cuneate to slightly orbicular, exhibiting a slight aureole around the point of attachment. Attachment is by means of a disc.

The distromatic nature of the frond readily distinguishes the plant from *P. nereocystis*. The cells are square or from once and a-half to twice as broad as high. The surface jelly is thick (Pl. XXI, figs. 15*a* and 15*b*).

The specimens examined show only antheridia, and from this it is supposed that the plant is diœcious. The antheridia are found towards the tip of the frond and along the edges. They evidently spread gradually over the whole frond.

The antheridia form the chief characteristic of the species. The antherozoids are arranged in groups such as are found in no other species, and exhibit a marked regularity in the division. As in all species of *Porphyra*, the antheridium-mother-cell, to produce the antheridia, undergoes a cruciate division perpendicular to the surface of the frond. The first reproductive division (in the antheridium) is parallel to the surface of the frond. This is followed by a cruciate division perpendicular to the surface of the frond, after which another parallel and another cruciate division take place, thus giving rise to sixty-four antherozoids in four tiers of sixteen each (Pl. XXI, figs. 16, 17*a*, 17*b*. Pl. XXII, fig. 27).

When the antheridia are fully ripe, the jelly-walls between the antherozoids dissolve before the surface jelly goes into dissolution, so that the antherozoids are arranged more or less irregularly in rectangular groups.

Habitat.—On rocks. Zone?

Distribution.—As yet reported from but a single locality on the Pacific Coast (36° 45′ N. lat.).

Locality.—Carmel Bay, Monterey County, California (Mrs. J. M. Weeks!).

V. ECONOMIC USES.

It is generally known that in Europe certain seaweeds are used as food and even sold in the markets, under the name of "laver." One of these seaweeds is a species of *Porphyra (P. laciniata)*. Kjellman (1897) devotes a part of his paper to a highly interesting account of the use of *Porphyra* species in Japan. From this it was learned that not only are the plants collected, but they are even cultivated, and after harvesting are carefully cleaned and prepared for market.

This led the writer to start on a voyage of discovery through Chinatown, San Francisco, to find out, if possible, whether the Chinese also use the species of *Porphyra* which occur in such abundance on our coast. It was found that formerly the Chinese used to depend altogether on *Porphyra* imported from Japan, but that now *P. perforata* and *P. nereocystis* are gathered in large quantities, especially at Monterey, and are partly consumed here and partly exported to China. The Chinese understand that there is some difference between the two species, having different names for them, and seemingly esteeming *P. nereocystis* more highly than *P. perforata*, at least the latter costs only about one-fifth the price of the former. Perhaps the distinction is identical with the distinction made by the Chinese between the plants collected from rocks and plants collected from *Nereocystis*.

Othes species of *Porphyra*, as yet undetermined, are sold in Chinatown in flat, round, purple-colored cakes, about one foot in diameter and from one-half to one-fourth of an inch thick. They are exported from Japan for Chinese use. This product forms a sharp contrast with "Asakusa Nori," also made of *Porphyra* sp. and exclusively used by Japanese. This is sold in bundles of ten sheets each, each sheet being about eight inches square, exceedingly thin, and light brown in color. While the latter are scrupulously clean and freed from all foreign algæ and animal matter, the product used by the Chinese contains other algæ as well as numerous molluscs.

The Indians of the slopes of the Pacific Coast also use *Porphyra* as a food, the tribes making yearly trips to the seashore to collect it, along with other economic seaweeds.

VI. Methods.

On account of the fact that in most of the species of *Porphyra* examined the jelly makes up a large part of the frond, considerable difficulty was encountered before the writer finally succeeded in preparing the necessary number of mounts. The specimens collected fresh were either dried or killed on the shore in Flemming's mixture, strong and dilute, one per cent. chromic acid, one per cent., two per cent. and five per cent. chrome alum, Wilson's corrosive sublimate, one per cent. formalin and alcohol. Of all these, Wilson's solution, formalin and alcohol, gave the best results, the others either shrinking the specimens or failing to fix them. As soon as this point was settled, all except the latter three were discarded. In some cases, as in *P. naiadum*, formalin gave the best results; while in others, Wilson's solution was to be preferred.

The necessity of the salt of a heavy metal to fix the jelly became apparent when rough dried and herbarium specimens were examined. The first ones to be studied were dried specimens of *P. perforata* which, when soaked in water, resumed their original outward form. But when the same process was tried with others, especially with *P. variegata*, it was found that the jelly dissolved very readily, so that some means had to be resorted to by which the specimens might be preserved in good condition. At first, glycerin, twenty per cent. glycerin in water, and twenty per cent. alcoholic glycerin were tried, but without good result. Finally Professor Setchell suggested the use of a saturated aqueous solution of corrosive sublimate. When used cold, this answered all requirements in every case except that of *P. variegata*, where it was necessary to use a boiling saturated aqueous solution.

Of all species of which fresh material could be obtained sections of from three to five microns in thickness were

made on the Minot microtome. To prepare the material for this purpose, small parts of the frond of which a section was desired were taken from the Wilson's solution in which they had been left for about six hours, or from the formalin in which they had remained an indefinite time, and placed in ten per cent. alcohol from which they were dialyzed into 95 per cent. alcohol. The specimens remained in 95 per cent. alcohol for an indefinite time, but never less than twenty-four hours. The parts of the frond were then folded in small bundles of about one-fourth of an inch or less square and one-eighth of an inch in thickness; after which they were transferred for three hours to 100 per cent. alcohol. From this they were placed in a mixture of equal parts absolute alcohol and bergamot oil. In this they remained two hours, as they did in the bergamot oil to which they were transferred. From the bergamot oil they were passed into 45° paraffin, a mixture of equal parts of 45° and 54° paraffin, and 54° paraffin, being during that time placed in a water-bath which was kept at a constant temperature of 56° C. In each of these they remained from two to three hours.

After this they were imbedded in 54° paraffin and on cooling were ready for cutting. In most cases good results were obtained.

This method was extremely useful in determining the number of divisions in the antheridia, which in comparatively thick razor sections with pith was very difficult to do.

Razor sections were usually stained with eosin, while microtome sections were stained with acid fuchsin or with safranin. Of the two, the latter gave the better results.

In a single case, that of *P. naiadum*, a double stain, methyl-blue-safranin, was used with exceedingly gratifying results. The slides were passed from xylol into absolute alcohol, 95 per cent. alcohol, and a saturated alcoholic solution of safranin. Here they remained for about half an hour, after which the slides were placed for five minutes in 95 per cent. alcohol to which some drops of methyl-blue, enough to give the alcohol a deep blue tinge, had been

added. After this they were transferred to absolute alcohol, xylol, and finally mounted in Canada Balsam. On examination the slides showed that while the cushion-shaped base of *P. naiadum* stained a bright red, the tissue of the host-plant, *Phyllospadix*, was stained blue, as were the chromatophores and cell-contents of *P. naiadum*. In some cases, where the slides had remained longer than five minutes in the methyl-blue, the young fronds emerging from the cushion-shaped bases were stained purple.

The ribbon was fixed on the slides with albumen, and floated out in water and placed on the top of the water-bath where the slides remained till dry.

As regards mounting media, dilute glycerin jelly and Farrants' Medium gave excellent results. While the former can be used to great advantage in preparing surface views, the latter is to be preferred where razor sections are to be mounted. It appears to have a clearing action on the tissues. Great care must be taken to wash out all corrosive sublimate in alcohol to which some potassium iodide is added, while hardening preparatory to cutting, since even the slightest trace of mercuric chloride will after a time attack the mounting medium and destroy the preparation.

In conclusion, the author begs to acknowledge his indebtedness to Professor W. A. Setchell, at whose suggestion the work was undertaken, and whose advice has been of invaluable aid in compiling this paper. Thanks are also due Professors Rosenvinge, Kjellman and Farlow, Messrs. Bornet, Collins, Janczewski, de A. Saunders, Lawson, Gibbs and Nott, and Mrs. Weeks and Mrs. Snyder, for specimens received.

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LITERATURE AND EXSICCATÆ CITED.

1821. AGARDH, C. A. Species Algarum. Gryphiswaldiæ.

1824. ——Systema Algarum. Lund.

1828. ——Icones Algarum Europæarum. Leipzig.

1882. AGARDH, J. G. Till Algernes Systematik, Afd. 3, VI, Ulvaceæ. *Lund's Univ. Årsskrift*, Tome XIX. Lund.

1899. ——Analecta Algologica, Continuatio V. *Acta Reg. Soc. Physiogr. Lund.*, Tome X. Lund.

1891. ANDERSON, C. L. List of California Marine Algæ, with Notes. *Zoe*, Vol. II. San Francisco.

1892. ——Algæ of the Farallons. (In Blankinship and Keeler: "On the Natural History of the Farallon Islands"). *Zoe*, Vol. III. San Francisco.

1882. BERTHOLD, G. Die Bangiaceen des Golfes von Neapel und der angrenzenden Meeres-Abschnitte. *Fauna und Flora des Golfes von Neapel und der angrenzenden Meeres-Abschnitte, herausgegeben von der zoölogischen Station zu Neapel.* Leipzig.

1882. COLLINS, F. S. Notes on New England Algæ. *Bull. Torrey Bot. Club*, Vol. IX. New York.

1884. ——Notes on New England Algæ, IV. *Bull. Torrey Bot. Club*, Vol. XI. New York.

1890. DE TONI, J. B. Frammenti Algologici. *La Nuova Notarisia*, Ano I, No. 3. Padova.

1897. ——*Sylloge Algarum*, Vol. IV. Padova.

1881. FARLOW, W. G. Marine Algæ of New England. *Report U. S. Comm. Fish and Fisheries*, 1879. Washington.

1890–1891. FOSLIE, M. Contribution to Knowledge of the Marine Algæ of Norway, I and II. *Tromsö Museums Aarshefter*, XIII. Tromsö.

1846–51. HARVEY, W. H. Phycologica Brittanica. London.

1849. ——British Marine Algæ. London.

1858. ——Nereis Boreali-Americana, Part III. Washington.

1893. HOWE, M. A. A Month on the Shores of Monterey Bay. *Erythea*, Vol. I. Berkeley, California.

1900. HUS, H. T. A. Preliminary Notes on West Coast Porphyras. *Zoe*, Vol. V. San Francisco.

1873. JANCZEWSKI, E. DE. Études Anatomiques sur les Porphyra. *Ann. Sci. nat.* (Bot.), 5e Sér., Tome XVII. Paris.

1883. KJELLMAN, F. R. The Algæ of the Arctic Sea. *K. Svenska Vet.-Akad. Handl.*, Band 20, No. 5. Stockholm.

1889. ——Om Beringhafvets Algflora. *K. Sv. Vet.-Akad. Handl.*, Band 23, No. 8. Stockholm.

1897. ——Japanska Arter af Slägtet Porphyra. *Bihang till K. Svenska Vetenskaps-Akademiens Handlingar*, Band 23, Afd. III, No. 4. Stockholm.

1864. LE JOLIS. Liste des Algues Marines de Cherbourg. *Mem. Soc. Imp. Sci. nat. de Cherbourg.*, Tome X. Paris.

1819. LYNGBYE, H. C. Tentamen Hydrophytologiæ Danicæ. Copenhagen.
1866. MARTENS, G. VON. Die Preussische Expedition nach Ost-Asien. Botanischer Theil. Die Tange. Berlin.
1897. MCCLATCHIE, A. J. Seedless Plants of Southern California. *Proc. Southern Calif. Acad.*, Vol. I. Los Angeles.
1893. ROSENVINGE, L. K. Grønlands Havalger. *Meddelelser om Grønland, III.* Kjobenhavn.
1894. ———Les Algues Marines du Grønland. *Ann. Sci. nat.* (Bot.) 7e Sér., Tome XIX. Paris.
1852. RUPRECHT. Neue oder unvol. bek. Pflanzen a. d. nördlichen Theile des Stillen Oceans. *Mém. Acad. de St. Petersb. sc. natur.*, Tome VII. St. Petersburg.
1882. SCHMITZ, F. Die Chromatophoren der Algen. Bonn.
1899. SETCHELL, W. A. Algæ of the Pribilof Islands. *The Fur Seals and Fur-Seal Islands of the North Pacific Ocean*, Part III. Washington.
1886. STRÖMFELT, H. F. G. Einige für die Wissenschaft neue Meeresalgen aus Island. *Bot. Centralblatt*, Bd. XXVI. Cassel.
1886. ———Om Algvegetationen vid Islands Kuster. Göteborg.
1870. SURINGAR, W. F. R. Algæ Japonicæ Musei botanici Lugduno-Batavi. Harlemi.
1878. THURET ET BORNET. Études Phycologiques. Paris.
1895–1901. COLLINS, HOLDEN & SETCHELL, Phycotheca Boreali-Americana, Fascl. I–XVII, and *A–B*. Malden, Mass.
1885–1896. HAUCK und RICHTER, Phycotheka Universalis, Fascl. I–XV. Leipzig.
1894–1901. TILDEN, American Algæ, Cent. I–V. Minneapolis.

EXPLANATION OF PLATE XX.

Figs. 1–6, and 11–14, drawn with Abbé's camera lucida, obj. Leitz $\frac{1}{4}$, ocular No. 3.

Figs. 7–10 drawn with Abbé's camera lucida, obj. Leitz $\frac{1}{4}$, ocular No. 1.

Porphyra leucosticta.

Fig. 1*a*. Surface view of vegetative part of frond.
Fig. 1*b*. Cross-section of vegetative part of frond.
Fig. 2*a*. Surface view of sporocarpic part of frond.
Fig. 2*b*. Cross-section of sporocarpic part of frond.
Fig. 3*a*. Surface view of antheridial part of frond.
Fig. 3*b*. Cross-section of antheridial part of frond.

Porphyra perforata.

Fig. 4*a*. Surface view of vegetative part of frond.
Fig. 4*b*. Cross-section of vegetative part of frond.
Fig. 5*a*. Surface view of sporocarpic part of frond.
Fig. 5*b*. Cross-section of sporocarpic part of frond.
Fig. 6. Cross-section of antheridial part of frond.
Figs. 7–10. Cells of the base, showing hypha-like projections; after treatment with Schulze's Macerating Fluid.

Porphyra nereocystis.

Fig. 11*a*. Surface view of cells of vegetative and sporocarpic part of frond.
Fig. 11*b*. Cross-section of vegetative part of frond.
Fig. 11*c*. Cross-section of sporocarpic part of frond.
Fig. 12. Cross-section of antheridial part of frond.

Porphyra amplissima.

Fig. 13*a*. Surface view of antheridial and sporocarpic part of frond.
Fig. 13*b*. Cross-section of antheridial and sporocarpic part of frond.

Porphyra miniata f. *cuneiformis.*

Fig. 14. Cross-section of antheridial and sporocarpic part of frond.

EXPLANATION OF PLATE XXI.

Figs. 15–18, and 20–22, drawn with Abbé's camera lucida, obj. Leitz ½, ocular No. 3.

Fig. 19 drawn with Abbé's camera lucida, obj. Leitz ⅔, ocular No. 3.

Porphyra occidentalis.

Fig. 15*a*. Surface view of vegetative part of frond.
Fig. 15*b*. Cross-section of vegetative part of frond.
Fig. 16. Cross-section of antheridial part of frond, showing stages in the development of the antheridia.
Fig. 17*a*. Surface view of antheridial part of frond.
Fig. 17*b*. Cross-section of antheridial part of frond.

Porphyra variegata.

Fig. 18. Cross-section of vegetative and sporocarpic part of frond.

Porphyra naiadum.

Fig. 19. Cross-section of blade of *Zostera* and of base of frond of *P. naiadum.*
Fig. 20*a*. Surface view of vegetative part of frond.
Fig. 20*b*. Cross-section of vegetative part of frond.
Fig. 21*a*. Surface view of sporocarpic part of frond.
Fig. 21*b*. Cross-section of sporocarpic part of frond.
Fig. 22. Cross-section of young frond with base.

EXPLANATION OF PLATE XXII.

Diagrams illustrating Formation of Reproductive Bodies.

Fig. 23. Cube, representing cell of *Porphyra* sp., which has undergone a cruciate division perpendicular to the surface of the frond. Formula for sporocarps, $4\left(\frac{a}{2}, \frac{b}{2}, c\right)$.

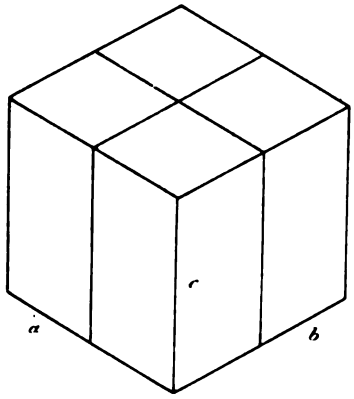
Fig. 24. The same, after undergoing a division parallel to the surface of the frond in each segment. Formula for sporocarps, $8\left(\frac{a}{2}, \frac{b}{2}, \frac{c}{2}\right)$.

Fig. 25. The same after undergoing another cruciate division perpendicular to the surface of the frond in all segments. Formula for sporocarps, $32\left(\frac{a}{4}, \frac{b}{4}, \frac{c}{2}\right)$; for antheridia, $8\left(\frac{\frac{1}{2}a}{2}, \frac{\frac{1}{2}b}{2}, \frac{c}{2}\right)$.

Fig. 26. The same after undergoing another division parallel to the surface of the frond in all segments. Formula for sporocarps, $64\left(\frac{a}{4}, \frac{b}{4}, \frac{c}{4}\right)$; for antheridia, $16\left(\frac{\frac{1}{2}a}{2}, \frac{\frac{1}{2}b}{2}, \frac{c}{4}\right)$.

Fig. 27. The same after undergoing another cruciate division perpendicular to the surface of the frond in all segments. Formula for antheridia, $64\left(\frac{\frac{1}{2}a}{4}, \frac{\frac{1}{2}b}{4}, \frac{c}{4}\right)$.

Fig. 28. The same after undergoing another division parallel to the surface of the frond in all segments. Formula for antheridia, $128\left(\frac{\frac{1}{2}a}{4}, \frac{\frac{1}{2}b}{4}, \frac{c}{8}\right)$.



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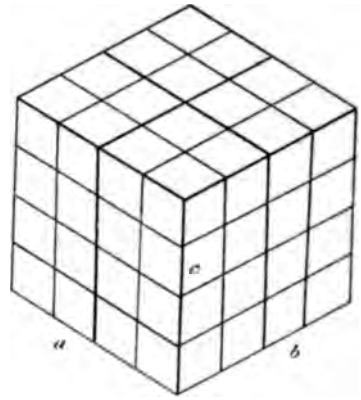

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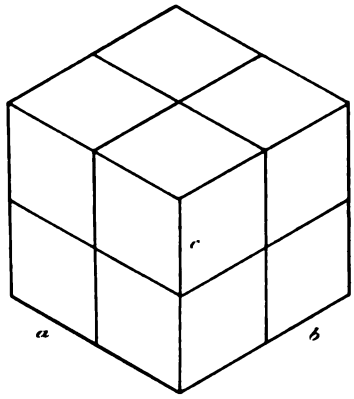

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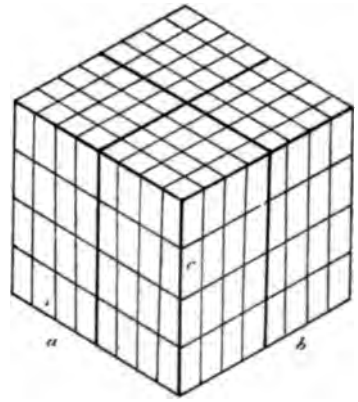

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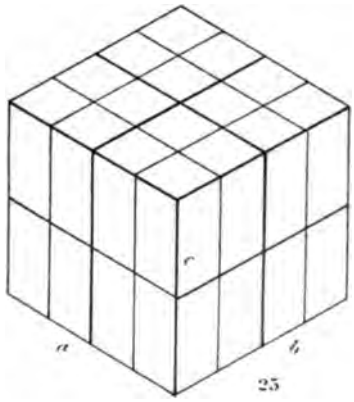

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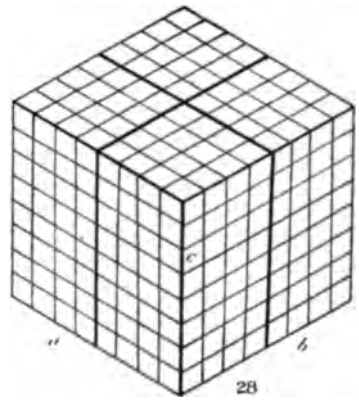

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BY

ALICE EASTWOOD

Curator of the Department of Botany

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SOME NEW SPECIES OF PACIFIC COAST RIBES.

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PLATES XXIII AND XXIV.

THE genus *Ribes* is represented on the Pacific Coast by four subgenera, which include an indefinite number of species and forms.

A recent attempt on the part of the author to identify the unnamed specimens of this genus, which had for years been accumulating in the Herbarium of the California Academy of Sciences, has brought to light the following species which seem to be undescribed.

The first six belong to the suborder Ribesia; of these, numbers one and two fall into the group typified by *Ribes sanguineum* Pursh, number three into that typified by *R. malvaceum* Smith; numbers four, five and six into that typified by *R. nevadense* Kellogg. The last three belong to the suborder Grossularia.

Some botanists might include the first six under *Ribes sanguineum*, the last three under *R. menziesii*. This aggregation may satisfy the amateur to whom generic differences are sufficient, but the real student desiring to learn the truth regarding a genus will find it a source of great confusion, and altogether unsatisfactory.

While it is to be kept in mind that nature knows no boundaries, and that orders, genera, and species are divisions made by man for his own convenience, yet these methods of classification have a scientific value beyond that of pure utilitarianism, and ought to show as far as possible the life-history of a group of related plants and of the entire plant world, when the knowledge of man makes it possible.

1. Ribes brandegei, sp. nov.

Plate XXIII, Figs. 1*a* and 1*b*.

Shrub with erect, branching stems unarmed; older bark dull brown; young bark glossy, not shreddy. Leaves three-lobed, reniform, 3–4 mm. long, and of about equal width; upper surface sparsely pubescent with fine, silky hairs, mostly on the veins, and with scattered sessile or shortly stipitate glands; lower surface pale green, with appressed, glandular hairs, veins at base villous; margin incised-dentate, glandular-ciliate; petioles generally shorter than the blades, glabrous or clothed with a fine pubescence under the gland-tipped hairs; stipular dilation 3 mm. broad, fringed with uneven, glandular hairs. Inflorescence racemose, erect in flower, the peduncles equalling or longer than the flowering portion, generally surpassing the leaves; pubescence as on the petioles; bracts foliaceous, oblanceolate to obovate, acuminate, incised, glandular-ciliate; flowers three to ten, on slender, erect pedicels which later become as long as the flower. Calyx rose-color, 8 mm. long, pubescent on the outside, glandular at base, puberulent within; divisions as long as the tube, oblanceolate to obovate, cucullate at summit, 2 mm. wide. Petals white, half as long as the calyx divisions, 1.5 mm. wide, orbicular-spatulate, on short claws. Stamens with slender filaments, 1.5 mm. long; anthers oblong, tipped with a blunt mucro. Style two-cleft at apex, with the stigmas broad. Berry glabrous, globular.

This species is related to *Ribes sanguineum* Pursh from which it differs in the pubescence, inflorescence, and shape of floral organs, as can be seen by the figures.

Collected by Mr. T. S. Brandegee, in whose honor the author takes pleasure in naming it, first at Sierra de Laguna, Lower California, January 25, 1890, later in the mountains of the Cape Region, March 26, 1892.

2. Ribes scuphami, sp. nov.

Plate XXIII, Figs. 2*a* and 2*b*.

Shrub with the upper bark reddish, shreddy, puberulent, unarmed. Leaves orbicular, three- to five-lobed, truncate to reniform at base, 2–5 cm. wide, about as long, unevenly dentate; upper surface pubescent with crisp, spreading hairs; lower, canescent with matted hairs; stipular dilation of the petiole broad, glandular, and tomentose, fringed with glandular hairs; petioles about as long as the blades, with pubescence like the stipules. Racemes numerous at the ends of the branches, 9 cm. long, slender, when flowering erect on peduncles which are shorter than the leaves; bracts oblanceolate, red, glandular, 8 mm. long, denticulate at apex; pedicels filiform, erect, a little longer than the bracts. Flowers subtended by two small, red bracteoles which are soon deciduous. Calyx rose-color, with tube 5 mm. long, divisions

linear-oblong, 7 mm. long. Petals white turning reddish, oblanceolate, cuneate, 4 mm. long. Stamens a little shorter than the petals; anthers globular. Ovary sparingly pubescent, and with scattered, stipitate glands.

This is nearest to *Ribes sanguineum* Pursh. It differs especially in having the racemes erect in flower, also in the more slender flowers with narrowed divisions. This species is the most beautiful of all belonging to the group of which *R. sanguineum* is the type.

It was collected on Smith River, Del Norte County, California, by Major J. R. Scupham, May, 1898. It is a pleasure to name this plant in honor of one who has brought many interesting plants to the herbarium of the California Academy of Sciences from little explored parts of California.

3. Ribes indecorum, sp. nov.

PLATE XXIII, FIGS. *3a* AND *3b*.

Shrub with erect stems, having dark brown, shreddy bark on the older growth, the younger parts tomentose and glandular. Leaves three-lobed, 2–4 cm. long, 2–3 cm. wide, finely rugose on the upper surface, clothed with stipitate glands, and a fine, sparse, silky pubescence; lower surface white with a felt-like tomentum, and with a few gland-tipped hairs on the veins; margins irregularly, doubly crenate; petioles stout, shorter than or equalling the blades, glandular and tomentose, the stipular dilation (as wide on each side as the petiole) fringed on the margin with uneven, gland-tipped hairs. Inflorescence racemose, spreading or pendent, in fruit surpassing the leaves; flowers sessile but erect; peduncles short; bracts foliaceous, almost equalling the flowers, lanceolate, 6 mm. long, 2 mm. wide, with the margins fringed with long, gland-tipped hairs. Peduncles stout, glandular, and tomentose. Flowers at base subtended by two membranous, glandular, and tomentose bracteoles; calyx-tube more than twice as long as the broad, rounded divisions; these tomentose and glandular on both sides, almost 2 mm. wide; petals orbicular, reniform, 1 mm. wide, crenulate, on very short and broad claws. Stamens as long as the petals, on stout, short, deltoid filaments; anthers .75 mm. long, longer than the filaments. Style stout, hairy at base, two-cleft at apex, with broad, yellow stigmas; ovary tomentose and somewhat glandular.

Collected by the author at Cajon Heights, near San Diego, California, March 14, 1891. There is also a specimen in the Herbarium of the California Academy of Sciences collected by Dr. George Thurber at San Pasqual, San Diego County. It is labeled *Ribes sanguineum*, No. 606.

Ribes indecorum is nearest to *Ribes malvaceum* but differs most noticeably in the much smaller and sessile flowers. The floral organs, too, are not the same.

4. Ribes ascendens, sp. nov.

PLATE XXIII, FIGS. 4*a* AND 4*b*.

Erect shrub unarmed, with gray-brown bark on older stems, younger stems paler and shreddy. Leaves three- to five-lobed, orbicular, reniform, 3-6 cm. wide, 2-5 cm. long, crenate-dentate, almost glabrous on the upper surface, the lower clothed with fine, spreading pubescence; petioles equalling or shorter than the blades, glandular; stipular dilation narrow, fringed with long, gland-tipped hairs. Peduncles generally surpassing the leaves, at first erect, later nodding, glandular-pubescent; flowers crowded at the summit of the peduncle, which is naked for more than half its length; bracts oblanceolate, rounded at apex, 7 mm. long, 2 mm. wide, with gland-tipped hairs on the surface and margin; pedicels half as long as the bracts, lengthening with age, and recurving upwards, so that the berries are erect. Flowers subtended by two membranous bracteoles which are soon deciduous. Calyx open-campanulate, rose-color, the tube about half as long as the divisions; these ovate, obtuse, 3.5 mm. long, 2.5 mm. wide, slightly pubescent. Petals white, orbicular, narrowed to a short, broad claw, 2 mm. wide. Stamens not equalling the petals, filaments linear, anthers oblong. Ovary clothed with gland-tipped hairs. Berry veiny, sparingly glandular, becoming 7 mm. or more in diameter.

This species is near *R. nevadense* Kellogg, but the racemes are ascending when in flower. The floral organs also differ in shape.

The type was collected by the author at Millwood, (Sequoia Mills) Fresno County, California, in flower, May 4, 1895; in fruit, July 18, 1893. There are specimens from General Grant Grove in the same vicinity, and from Coburn's Mills in Fresno County, collected by T. S. Brandegee; the former, July, 1892, the latter, May 29 (year not given).

4*a*. *Ribes ascendens jasperæ*, var. nov. (Plate XXIV, fig. 5). This is similar to the species in general appearance, but the sepals and petals are more orbicular, the filaments are shorter, and the anthers broader and more orbicular. It is named in honor of Mrs. William Jasper, who sent the specimen from San Emidio Cañon, Kern County, California, May, 1895.

5. Ribes hittellianum, sp. nov.

PLATE XXIV, FIGS. 6*a* AND 6*b*.

Erect shrub with spreading branches, 2–3 feet high; bark smooth, unarmed, gray-brown, shreddy on the younger branches. Leaves three- to five-lobed, orbicular, reniform or truncate at base, 3–4 cm. wide, 2–3 cm. long, irregularly dentate and somewhat revolute, rugulose veiny, glabrous on both sides but with some scattered glands on the lower; petioles about as long as the blades, sparingly tomentose and glandular; stipular dilation broad, truncate, membranous, as wide on each side as the petiole. Racemes 1–2 cm. long, at first erect, later nodding, but with the pedicels erect; flowers three to eight, crowded, subtended by deciduous bracteoles. Calyx tubular-campanulate, with the tube 1 mm. long, the divisions rose-color, oblong, obtuse, 4 mm. long, 2 mm. wide. Petals white, narrowly oblong, three-fourths as long as the sepals and one-half as wide. Stamens with subulate filaments, half as long as the sepals; anthers orbicular. Stigmas two, capitate; ovary and immature fruit clothed with stipitate glands.

This species belongs in the group of which *R. nevadense* Kellogg is the type. It differs from that species in the inflorescence and the shape of the floral organs.

Collected near the head-waters of Cañon Creek, Trinity County, California, not far from Twin Lakes, July 9, 1901, and named in honor of Mr. Carlos T. Hittell, one of the party on a trip to these little known mountains.

6. Ribes glaucescens, sp. nov.

PLATE XXIV, FIGS. 7*a* AND 7*b*.

Unarmed shrub with older bark gray-brown, younger bark bright brown glossy, shreddy. Leaves three-lobed, orbicular-reniform, about 3 cm. long, 3–5 cm. wide, irregularly dentate, glabrous except for some minute glands on the lower surface, glaucescent, paler on the lower surface; petioles about as long as the blade, minutely puberulent, with the stipular dilation on each side narrower than the petiole, and sparingly fringed with glandular hairs. Inflorescence in fruit spreading or erect, generally shorter than the leaves, rather loosely flowered with from five to ten flowers; peduncles as long as the raceme, striate, puberulent; pedicels slender, becoming 5 mm. long, shorter than the brown, membranous, gland-tipped bracts. Flowers subtended by two small, deciduous, reddish bracteoles. Calyx open-campanulate, with very short tube, and spreading divisions; these rose-color, oblanceolate, 4 mm. long, 1.5 mm. wide, glabrous. Petals white, spatulate two-thirds as long as the sepals, denticulate near the apex. Stamens half as long as the sepals, with suborbicular anthers and broad filaments. Ovary glabrous except for the scattered stipitate glands.

This species is related to *Ribes nevadense* Kellogg from which it differs in the glaucous color of its foliage, the racemes erect in fruit, and the shape of the floral organs. It was collected by the author on Mount Shasta, August 13, 1893. No note was taken of the exact locality, but it must have been some place on the trail from Sisson.

7. **Ribes oligacanthum,** sp. nov.

PLATE XXIV, FIGS. 8*a* AND 8*b*.

Erect, branching shrub with the younger stems puberulent, older gray-brown, not shreddy, somewhat tortuous, unarmed, except for the simple or triple thorns at the leaf-axils; these often short and abortive. Leaves broadly ovate or orbicular, three- to five-lobed, 2–3 cm. wide and about as long, with margin laciniate-dentate, base truncate, but cuneate where it joins the petiole, glabrous; petioles slender, as long as or twice as long as the blades, glabrous or puberulent; stipular dilation twice as wide as the petiole, ciliate with white, silky hairs. Peduncles filiform, one- to three-flowered, 1–2 cm. long; pedicels almost as long, together becoming 4 cm. long, slightly puberulent; bracts broadly ovate to orbicular, acuminate, three-angled, clasping, reflexed. Calyx 12 mm. long, the linear, acute segments more than twice as long as the tube, 2 mm. wide, apparently white, veined with parallel veins that branch near the apex, puberulent or glabrous. Petals involute, obovate when spread out, irregularly denticulate at apex, 3 mm. long, veins palmate. Filaments almost twice as long as the petals, dilated at base; anthers oblong-ovate, cordate at base, tipped with a recurved mucro. Style divided two-thirds of its length, slightly exserted. Young fruit puberulent, clothed with a few long spines (about ten) each about 5 mm. long.

Related to *Ribes californicum* Hook. & Arn. from which it differs in the sparsely spinous fruit, the glabrous leaves, the lax inflorescence, and the shape of the floral organs.

Collected by the author on the road between Jolon and King City, in Monterey County, California, near Mansfield's Ranch, ten miles from King City, May, 1897.

8. **Ribes sericeum,** sp. nov.

PLATE XXIV, FIGS. 9*a*-9*f*.

Erect, branching shrub, several feet high; stems clothed with numerous fine, weak, short prickles, which are gland-tipped on the young shoots, also with short, close, silky pubescence; axillary thorns three, orange-color, stout, united, the middle one longest, more than 1 cm. long, broadening at the

base, pubescent and glandular on the lower part, glabrous on the upper. Leaves thin, three- to five-lobed, broadly ovate-orbicular, reniform or truncate at base, 2-4 cm. long, not quite so wide, incised-crenate, clothed with fine, white, silky hairs which are appressed or spreading, also with fine gland-tipped hairs; petioles about as long as the blades, more glandular and more spreading-pilose, dilated only at the very base, and without the appearance of stipules. Peduncles one- to three-flowered, slender, erect, with pubescence like the petioles; pedicels about half as long; bracts orbicular or three-lobed, foliaceous; bractlets similar but smaller. Flowers 2 cm. long, open-campanulate in the bud. Calyx with the divisions at length reflexed, longer than the tube and the ovary, oblong, purplish red, greenish near the apex, softly silky villous on both sides; tube campanulate, veined, slightly glandular at base. Petals white, 5 mm. long, involute, erose along the almost truncate apex. Stamens with filiform, purple filaments, exserted beyond the sepals in the opening flower, and also beyond the pistil; anthers narrowly linear-oblong, almost 2 mm. long, obtuse. Pistil two-cleft for 2 mm. Ovary densely clothed with horizontally spreading fine, silky hairs mixed with some longer, glandular hairs, the glands purple. Fruit purple, clothed with short, weak bristles and scattered hairs. Some of the bristles retain the purple glands on the fruit.

Collected in flower by Mr. R. A. Plaskett, at Spruce Creek; also at Gorda, in flower and fruit. Collected by the author at Pacific Valley, with immature fruit. *R. sericeum* flowers in December and January and fruits in June. At Point Sur specimens were collected by the author in June, 1893, with very large, pear-shaped fruit, almost 4 cm. long, and specimens with globular fruit were collected at about the same time at Slate's Hot Springs. All these localities are on the coast of Monterey County, California, at the base of the Santa Lucia Mountains, and the range extends from south of Point Gorda to north of Point Sur.

Ribes sericeum is related to *R. subvestitum* Hook & Arn. but it has different leaves, different pubescence, and the floral organs are not the same.

8*a*. *Ribes sericeum viridescens*, var. nov. The variety is similar to the type, but the flowers are smaller and greenish, the leaves are more densely clothed with silky white hairs, and are more orbicular-reniform. The peduncles in the specimens examined all have single flowers.

This variety was collected by R. A. Plaskett at Gorda, Monterey County, California, January, 1898.

9. Ribes hystrix, sp. nov.

PLATE XXIV, FIGS. 10*a*–10*d*.

Shrub several feet high, with light brown, tortuous branches, minutely pubescent and thickly beset with stout, rigid, horizontal, yellow prickles, some gland-tipped, generally small on the new growth, and increasing in size with age; axillary thorns triple, stout, distinct at base, middle one longest, becoming 15 mm. long, lower part pubescent, upper, glabrous. Leaves thin, three-lobed or some five-lobed, with the basal lobes small, 2–4 cm. wide, orbicular-reniform, incisely dentate, minutely pubescent and dotted with sessile glands on the lower surface, almost glabrous on the upper; petioles about as long as the blade, tomentose and slightly glandular. Peduncles one- to three-flowered, 1–2 cm. long, ascending, slender, sparingly pilose and clothed with gland-tipped hairs; pedicels less than half as long, occasionally longer; bracts orbicular or lobed, clasping, acuminate to obtuse. Calyx pubescent and glandular; tube a little longer than the ovary; divisions 1 cm. long, surpassing the rest of the flower when reflexed, 3 mm. wide, lower part purple, near the apex greenish, obtuse. Petals white, broadly obovate when spread out, acute, narrowed to a short claw, involute, 4 mm. long. Stamens with broad filaments dilated at base, as broad and long as the anthers; these about 3 mm. long, sagittate at base, tipped with a blunt mucro. Styles surpassing the sepals in the opening flower, divided about half; stigmas small, capitate; ovary globular, tomentose, and densely clothed with purplish bristles, some near the calyx gland-tipped. Fruit purple, more or less densely clothed with stiff, spreading prickles, 2–5 mm. long.

This species is nearest to *R. menziesii* Pursh but differs in the glandular pubescence, the shape and texture of the leaves, the size of the flower, and shape of the parts.

Collected in flower by Mr. R. A. Plaskett, at Gorda, Santa Lucia Mountains, California, December, 1897. The fruiting specimens were collected by the author at Pacific Valley, in the same vicinity, May, 1897, and June, 1893.

KEY TO THE SPECIES OF PACIFIC COAST RIBES.

The following key, made by the author for convenience in looking up the described species of Pacific Coast *Ribes*, is appended, as it may be useful to others under the same circumstances.

Chrysobotrya SPACH. GOLDEN CURRANT.

Calyx with long tube; flowers yellow, berry smooth, leaves convolute in bud; stems without thorns or prickles.

1. *R. tenuiflorum* LINDL., Bot. Reg., Vol. XV, Tab. 1274. Oregon and California.
2. *R. aureum* PURSH, Fl. Am. Sept., Vol. I, p. 163. Oregon and Washington.

Ribesia BERLANDIER. WILD CURRANT.

Calyx-tube cylindrical to rotate; berries smooth; leaves plicate in bud; stems without thorns or prickles; flowers in racemes.

Calyx-tube cylindrical.

3. *R. bracteosum* DOUGL., Hook. Fl. Bor. Am., Vol. I, p. 233. Northern California to Alaska.
4. *R. cereum* DOUGL., Bot. Reg., Vol. XV, Tab. 1263. California to British Columbia.
5. *R. viscosissimum* PURSH, Fl. Am. Sept., Vol. I, p. 163. Sierra Nevada of California to British Columbia.
6. *R. sanguineum* PURSH, Fl. Am. Sept., Vol. I, p. 164. Northern California to British Columbia.
7. *R. scuphami*, sp. nov. Del Norte County, California.
8. *R. brandegei*, sp. nov. Lower California.
9. *R. glutinosum* BENTH., Trans. Hort. Soc., Ser. II, Vol. I, p. 476. Coast Mountains of California.
10. *R. malvaceum* SMITH, Rees. Cycl., Vol. XXX. Coast Mountains of California.
11. *R. palmeri* VASEY & ROSE, Proc. U. S. Nat. Mus., Vol. XI, p. 529. Lower California.
12. *R. indecorum*, sp. nov. San Diego County, California.
13. *R. nevadense* KELLOGG, Proc. Cal. Acad. Sci., Vol. I, 1855, p. 65 = (*R. sanguineum variegatum* WATS., Bot. King's Exped. 100th Par.) Sierra Nevada of Central California.
14. *R. ascendens*, sp. nov. Sierra Nevada of Central California.
15. *R. hittellianum*, sp. nov. Trinity County, California.
16. *R. glaucescens*, sp. nov. Mount Shasta, California.
17. *R. viburnifolium* GRAY, Proc. Am. Acad., Vol. XVII, p. 202. Lower California, and islands off the coast of Santa Barbara, California.

Calyx rotate or saucer-shaped.

18. *R. prostratum* L'HÉRIT, Strip. Nov. 3, Tab. 2. British Columbia and northward.
19. *R. hudsonianum* RICHARDS., Frank. Journ. 2nd Ed., App., p. 6. British Columbia and northward.
20. *R. migratorium* SUKSDORF, Deutsche Bot. Monats., Bd. XVIII, p. 86. Washington.
21. *R. laxiflorum* PURSH, Fl. Am. Sept., Vol. II, p. 731. Washington and northward.
22. *R. ciliosum* HOWELL, Fl. N. W. Am., p. 208. Mount Hood, Oregon.
23. *R. erythrocarpum* COVILLE & LEIBERG, Proc. Biol. Soc. Wash., Vol. X, p. 132. Crater Lake, Oregon.

Grossularia A. RICHARD. GOOSEBERRY.

Stems thorny under the fascicles, generally prickly besides; leaves plicate in bud; flower solitary, in corymbs, or racemes.

Berry prickly or glandular.

24. *R. menziesii* PURSH, Fl. Am. Sept., Vol. II, p. 732 (= *R. ferox* SMITH, Rees. Cycl., Vol. XXX. [Index Kewensis]). Coast Mountains of California.
25. *R. hystrix*, sp. nov. Santa Lucia Mountains, California.
26. *R. californicum* H. & A., Bot. Beech. Voy., p. 346. Coast Mountains of California.
27. *R. occidentale* H. & A., Bot. Beech. Voy., p. 346. Coast Mountains of California. A doubtful species.
28. *R. oligacanthum*, sp. nov. Santa Lucia Mountains, California.
29. *R. victoris* GREENE, Pittonia, Vol. I, p. 224. Coast Mountains of California.
30. *R. mariposanum* CONGDON, Erythea, Vol. VII, p. 183. Mariposa County, California.
31. *R. subvestitum* H. & A., Bot. Beech. Voy., p. 346. Coast Mountains of California.
32. *R. sericeum*, sp. nov. Santa Lucia Mountains, California.
33. *R. lobbii* GRAY, Am. Nat., Vol. X, p. 274. Northern California to Vancouver.
34. *R. marshallii* GREENE, Pittonia, Vol. I, p. 31. Northern California to Oregon.
35. *R. amictum* GREENE, Pittonia, Vol. I, p. 69. Sierra Nevada of California.
36. *R. wilsonianum* GREENE, Erythea, Vol. III, p. 70. Mountains of Kern County, California.
37. *R. cruentum* GREENE, Pittonia, Vol. IV, p. 35. Coast Mountains of California to Oregon.
38. *R. aridum* GREENE, Pittonia, Vol. IV, p. 35. Sierra Nevada in Kern County, California.

39. *R. hesperium* McCLATCHIE, Erythea, Vol. II, p. 79. Los Angeles County, California.
40. *R. amarum* McCLATCHIE, Erythea, Vol. II, p. 79. Los Angeles County, California.
41. *R. montigenum* McCLATCHIE, Erythea, Vol. V, p. 38.
42. *R. lacustre* POIR, Encycl. Suppl. II, p. 856 (= *R. echinatum* DOUGL., Bot. Reg., sub. Tab. 1349 [Index Kewensis]). California, northward.
43. *R. lacustre* var. *molle* GRAY, Bot. Cal., Vol. I, p. 206. Sierra Nevada Mountains, northward.

Berry smooth.

44. *R. divaricatum* DOUGL., Trans. Hort. Soc., Vol. VII, p. 515 (= *R. villosum* NUTT., T. & G. Fl. N. Am., Vol. I, p. 547). California.
45. *R. oxyacanthoides* L., Sp. Pl., p. 201 (= *R. saxosum* HOOK., Fl. Bor. Am., Vol. I, p. 231 [Index Kewensis]). Sierra Nevada of California to British Columbia.
46. *R. gracile* MICHX., Fl. Am. Bor., Vol. I, p. 111. Oregon to British Columbia.
47. *R. niveum* LINDL., Bot. Reg., Vol. XX, Tab. 1692. Washington.
48. *R. leptanthum* GRAY, Mem. Am. Acad., N. Ser., Vol. IV, p. 53. Sierra Nevada of California.
49. *R. cognatum* GREENE, Pittonia, Vol. III, p. 115. Oregon.
50. *R. lasianthum* GREENE, Pittonia, Vol. III, p. 22. Sierra Nevada of California.
51. *R. velutinum* GREENE, Bull. Cal. Acad. Sci., Vol. I, No. 3, p. 83. California to northern Oregon.
52. *R. quercetorum* GREENE, Bull. Cal. Acad. Sci., Vol. I, No. 3, p. 83. Coast Mountains of California. This is the same as *R. leptanthum brachyanthum* GRAY, according to Greene.
53. *R. ambiguum* WATS., Proc. Am. Acad., Vol. XVIII, p. 193. Washington. This has been changed to *R. watsonianum* KOEHNE.
54. *R. montanum* HOWELL, Fl. N. W. Am., p. 210. Siskiyou Mountains, Oregon. This has been changed to *R. binominatum* HELLER.

Robsonia BERLANDIER.

Stems thorny; parts of the flower commonly four; calyx with erect lobes.

55. *R. speciosum* PURSH, Fl. Am. Sept., Vol. II, p. 731 (= *R. stamineum* SMITH, Rees. Cycl., Vol. XXX = *R. fuchsioides* BERL., Mem. Soc. Geneva, Vol. III, Pt. 2, p. 58.) Monterey, California, southward.

The references not being available, the following species could not be placed:—

56. *R. roezli* REGEL, Gartenfl., p. 226. Am. Bor. occ. [Index Kewensis].
57. *R. spæthianum* KOEHNE.

EXPLANATION OF PLATE XXIII.

Fig. 1. *Ribes brandegei*, sp. nov.
- *a.* Diagrammatic representation of the flower; × 5.
- *b.* Leaf, natural size.

Fig. 2. *Ribes scuphami*, sp. nov.
- *a.* Diagrammatic representation of the flower; × 5.
- *b.* Leaf, natural size.

Fig. 3. *Ribes indecorum*, sp. nov.
- *a.* Diagrammatic representation of the flower; × 5.
- *b.* Leaf, natural size.

Fig. 4. *Ribes ascendens*, sp. nov.
- *a.* Diagrammatic representation of the flower; × 5.
- *b.* Leaf, natural size.

Fig. 5. *Ribes ascendens jasperæ*, sp. et var. nov.

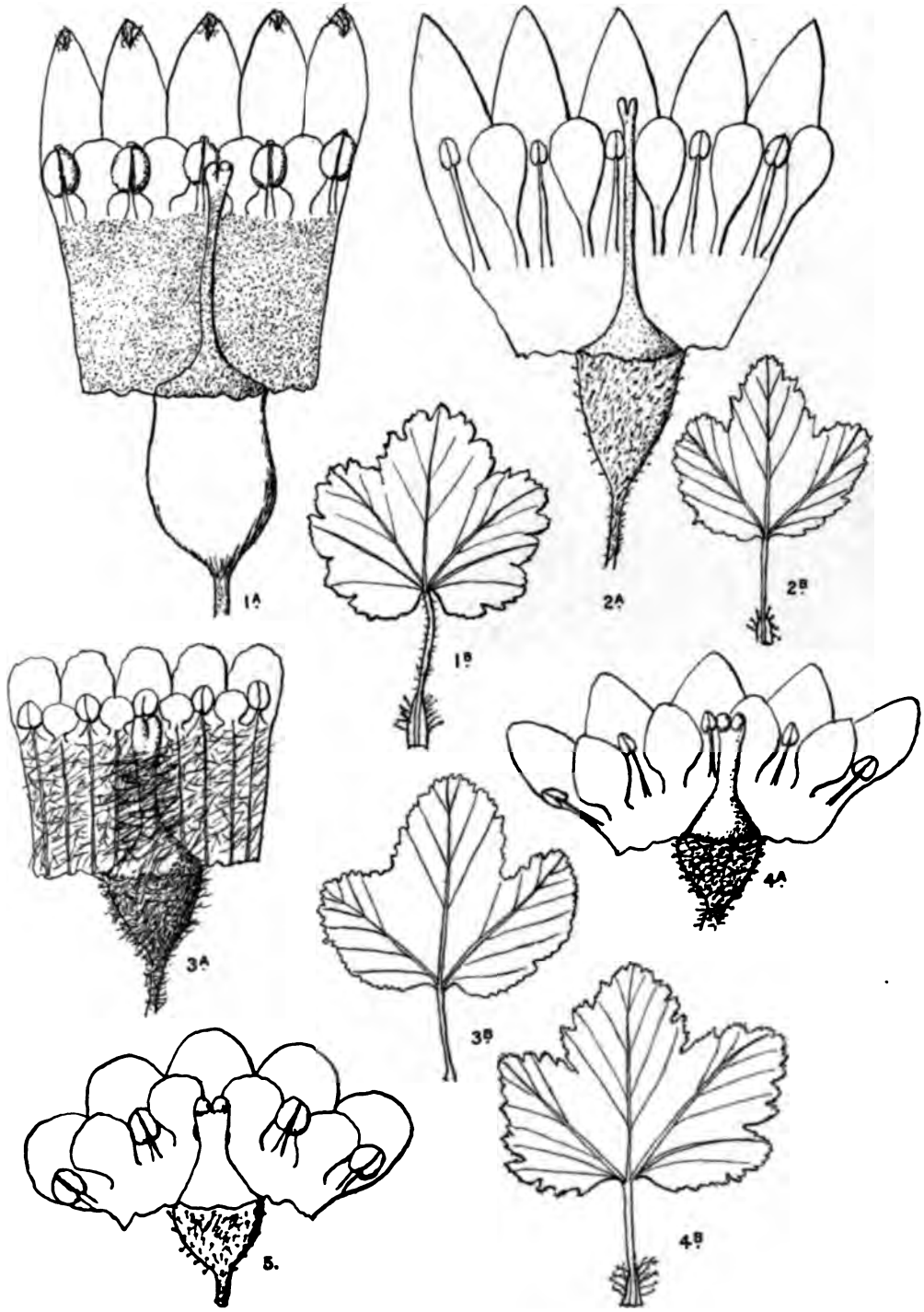

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FIG. 1. RIBES BRANDEGEI, SP. NOV.
FIG. 2. RIBES SCUPHAMI, SP. NOV.
FIG. 3. RIBES INDECORUM.
FIG. 4. RIBES ASCENDENS.
FIG. 5. RIBES ASCENDENS, JASPERÆ, SP. ET VAR. NOV.

EXPLANATION OF PLATE XXIV.

Fig. 6. *Ribes hitchellianum*, sp. nov.
- *a*. Diagrammatic representation of the flower; × 5.
- *b*. Leaf, natural size.

Fig. 7. *Ribes glaucescens*, sp. nov.
- *a*. Diagrammatic representation of the flower; × 5.
- *b*. Leaf, natural size.

Fig. 8. *Ribes oligacanthum*, sp. nov.
- *a*. Diagrammatic representation of the flower; × 5; dotted lines indicate the shape of the ovary and calyx-tube.
- *b*. Leaf, natural size.

Fig. 9. *Ribes sericeum*, sp. nov.
- *a*. Flower, natural size.
- *b*. Buds, natural size.
- *c*. Leaf, natural size.
- *d*. Petal, spread out; × 2.
- *e*. Petal as it appears in flower; × 2.
- *f*. Stamen; × 2.

Fig. 10. *Ribes hystrix*, sp. nov.
- *a*. Flower and bud, natural size.
- *b*. Leaf, natural size.
- *c*. Stamen; × 2.
- *d*. Petal; × 2.

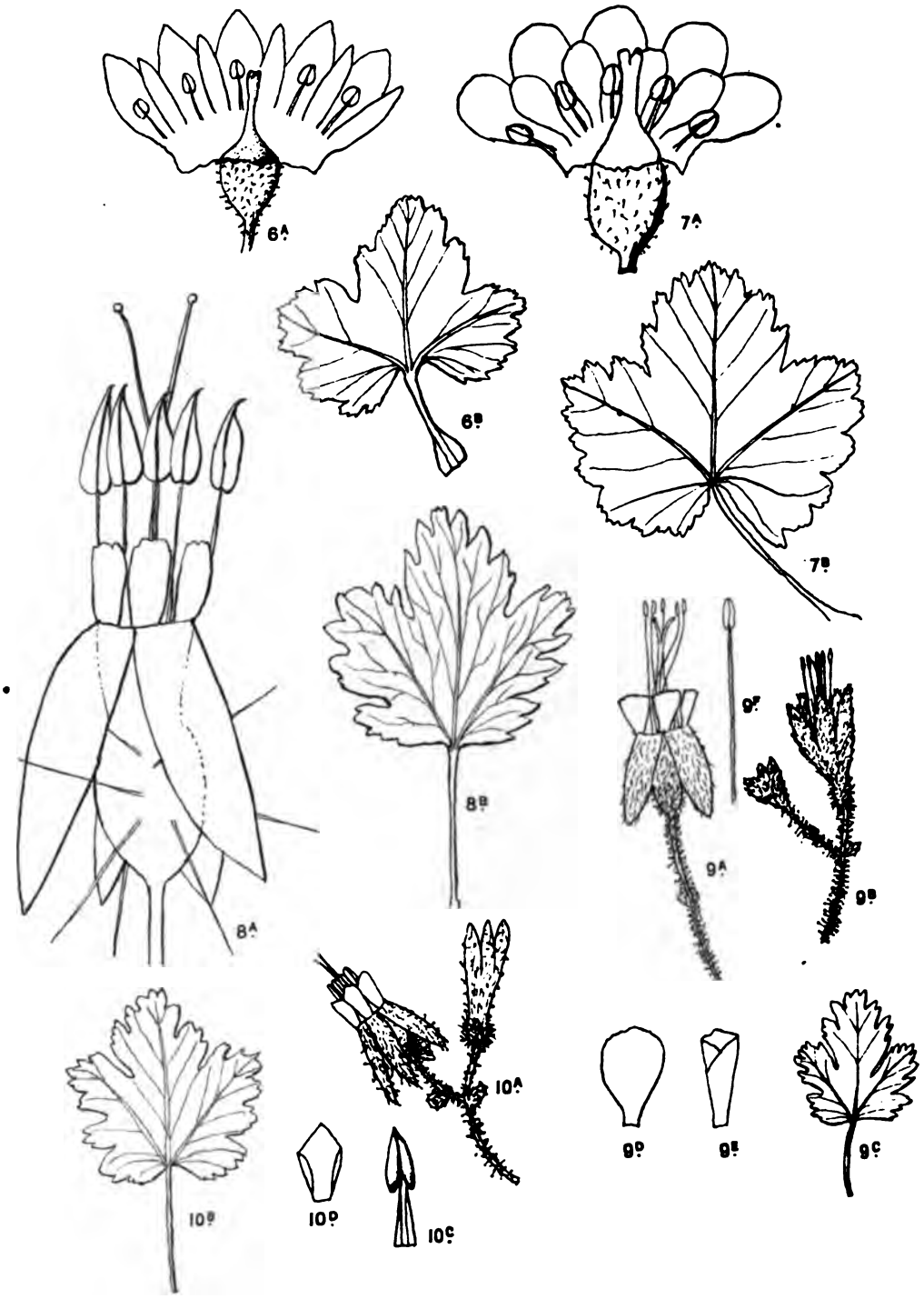

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FIG. 6. RIBES HITTELLIANUM, SP. NOV.
FIG. 7. RIBES GLAUCESCENS, SP. NOV.
FIG. 8. RIBES OLIGACANTHUM, SP. NOV.
FIG. 9. RIBES SERICEUM, SP. NOV.
FIG. 10. RIBES HYSTRIX, SP. NOV.

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Cell Studies

I. Spindle Formation in Agave

BY

W. J. V. OSTERHOUT

Assistant Professor of Botany in the University of California

WITH FOUR PLATES

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CELL STUDIES.[1]

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BY W. J. V. OSTERHOUT,

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I. INTRODUCTION AND TECHNIQUE.

UNDER this title I propose to publish the results of a series of investigations bearing on various cell questions. The especial attention which has been devoted to technique in these investigations makes it desirable to refer at the

[1] Contributions from the Botanical Laboratories of the University of California.

April 30, 1902.

outset to the methods employed. The particular point of technique which demands the most care in dealing with plant cells is fixation. The artifact caused by many fixing agents cannot be too carefully guarded against; it was deemed advisable therefore to test each fixative by observing its effect on living cells. This was accomplished by teasing out the living cells on the slide in the liquid expressed from surrounding tissues, and observing them while irrigation with the fixative was going on. By employing a Zeiss 2 mm. apochromat with comp. Oc. 6, 8 and 12, the effect of the fixative on the finer details of structure could be carefully followed. Some of the most highly recommended fixatives were seen to produce profound disturbances in the structure of the cytoplasm and were therefore modified in various ways in hope of better results. The fact that in different stages of development the cell reacts differently necessitated a separate test for each stage. The principal fixatives used are as follows:—

1. Flemming's strong chromic-osmic-acetic.
2. Flemming's strong chromic-osmic-acetic diluted with from one to twenty parts of water.
3. Flemming's strong chromic-osmic-acetic plus from one-tenth to one part of glacial acetic.
4. Flemming's strong chromic-osmic-acetic modified as follows:—

   Chromic 7 per cent. 8 c. c.
   Osmic 2 per cent. 2 c. c.
   Glacial acetic 0.5 c. c.
5. Flemming's strong chromic-osmic-acetic with the acetic omitted.
6. Hermann's platinic chloride-acetic-osmic.
7. The same diluted with from one to twenty parts of water.
8. Iridium chloride 1 per cent.
9. Iridium chloride plus 1 per cent. of glacial acetic.
10. Iridium chloride .5 per cent.
11. Iridium chloride .5 per cent. plus 1 per cent. of glacial acetic.

12. Palladium chloride 1 per cent.
13. Palladium chloride 1 per cent. plus 1 per cent. of glacial acetic.
14. Osmium chloride 1 per cent.
15. Osmium chloride 1 per cent. plus 1 per cent. of glacial acetic.
16. Platinum chloride 1 per cent.
17. Platinum chloride 1 per cent. plus 1 per cent. of glacial acetic.
18. Chromic 1 per cent.
19. Glacial acetic 1 per cent.
20. Potassium bichromate C. P. 3 gm.
    Glacial acetic 5 c. c.
    Distilled water 95 c. c.
21. Iodine, saturated solution in distilled water.
22. Potassium iodide 0.5 gm.
    Iodine 1. gm.
    Distilled water 1000 c. c.
23. Boveri's picro-acetic.
24. Wilson's sublimate-acetic.
25. Sublimate, saturated solution in alcohol.
26. Rawitz's picro-nitric.
27. Guignard's iron chloride-chromic-acetic.
28. Guignard's iron chloride-picric.
29. Merkel's platinic chloride-chromic.
30. Lindsay Johnson's bichromate-platinic chloride-osmic-acetic.
31. Von Rath's sublimate-picric-osmic.
32. Tellyesniczky's bichromate-acetic.
33. Zenker's bichromate-sodium sulphate-sublimate-acetic.
34. Kleinenberg's picro-sulphuric.
35. Carnoy's alcohol-chloroform-acetic.
36. Mann's sublimate-tannin-picric.
37. Mann's alcohol-sublimate-tannin-picric.
38. Rabl's sublimate-platinic chloride.
39. Keiser's sublimate-acetic.
40. Mayer's saltpetre-picric.

Flemming's strong mixture (1) proved decidedly the best in most cases; very fair results were obtained also with iridium chloride (8), platinum chloride (16), palladium chloride (11), and Flemming's strong mixture with an excess of chromic (4). Keiser's sublimate-acetic (39) gave good results in some cases.

Some objects could not be satisfactorily fixed in any of these. In such cases good results were usually obtained by the use of Flemming's strong mixture with the acetic omitted (5); where this failed resort was had to fixing in potassium iodide-iodine (22) and transferring after a longer or shorter time to Flemming's strong mixture.

In most cases fixatives were found which produced no visible change in the structure of the living cell so far as could be ascertained by careful observation with high powers during the application of the fixative. It was found, however, that material placed in some of these fixatives undergoes structural alterations after a few hours; in these cases the time of fixation was shortened accordingly.

During the formation of the spindle the cell is more sensitive (i. e., more apt to shrink or produce artifact) to the action of fixatives than at any other time; it is not quite so sensitive after the spindle is fully formed; during the anaphase it grows less and less so. Pollen-mother-cells are usually quite sensitive just after division into tetrads. Resting stages of the cell are the least sensitive of all, and the degree of sensitiveness seems to be in inverse ratio to the amount of protoplasm in the cell. The nucleus is always far more resistant than the cytoplasm.

At every step in the processes subsequent to fixing, the material was subjected to careful examination in order to guard against artifact. These observations showed that the points requiring most care are the washing out of the fixative and the infiltration with oil. Either of these processes if too prolonged will frequently affect the structure of the cytoplasm; they should therefore be carefully controlled.

The treatment of the material after fixing was as follows:

After being washed from two to eight hours in running water it was transferred to a dehydrator (for description of this see Lawson, 1898, Williams, 1899, or Osterhout, 1900). Dehydration in no case produced shrinkage when allowed to proceed with sufficient slowness. The material remained in the dehydrator until the strength of the alcohol in the upper part was equal to that in the lower. The relative strength of the alcohols may be ascertained as follows: Dip a pipette into the alcohol in the upper part and allow it to take up a few drops by capillary attraction; now dip the pipette into the other alcohol, if on diffusing out of the pipette it falls, it is weaker (on holding it up to the light the course of the diffusion current is easily seen). The material was transferred from the dehydrator to a mixture consisting of equal parts of 95 per cent. alcohol and alcohol from the dehydrator. After standing two hours a part of this alcohol was removed, mixed with an equal quantity of 95 per cent. alcohol, and the material transferred to this mixture. It then passed in succession through 95 per cent. alcohol (2–6 hours), absolute alcohol (2–6 hours), absolute alcohol and bergamot oil, equal parts (6–12 hours), and bergamot oil (3–6 hours). It was then placed on the top of the paraffin oven and allowed to warm up slowly, and was then transferred to bergamot oil and paraffin—equal parts—and allowed to remain on the top of the bath. After six to twelve hours it was placed within the oven, and after an hour transferred to paraffin 43° (12 hours), and finally to paraffin 52°, 60° or 72°.

Sections 1–5 μ thick were cut on the Minot wheel microtome, and fastened to the slide by the water-albumen method. For preliminary examination of ribbons Eisen's alcohol method (Eisen, 1897) proved exceedingly useful; the drying on the oven was omitted, thus effecting a great saving of time without any corresponding disadvantage (the sections were examined in xylene only, in which they do not tend to float off as they do when examined in water, without having been carefully dried on the oven). In view of the fact that the kinoplasmic fibres, and especially the

genetic fibres, are more difficult to stain than other constituents of the cell, search was made for a stain which should differentiate these structures sharply. Out of the extensive series of stains tested, gentian violet proved the best, especially when combined with safranin and orange G in the Flemming triple stain. Benda's iron-hæmatoxylin, used in combination with Bordeaux red, congo red, or ruthenium red, gives good results in the study of non-kinoplasmic elements. The use of gentian violet is attended with some difficulties; in many cases sections can be left but a short time in the violet, and it is then found that the absolute alcohol washes out the violet too rapidly. This may be avoided by dissolving enough gentian violet in the absolute alcohol to make a saturated solution. The washing out of the violet can then be done entirely by means of clove or anilin oil (either pure or diluted with xylol), and this in many cases is of great advantage. Dipping the slides from two to twenty seconds in dilute potassium-iodide-iodine solution just before transferring to absolute alcohol was found in many cases to give a sharper differentiation of all the structures stained by the violet.

The intense illumination necessary for work with high oculars was secured by using a Welsbach light, and interposing a glass globe (about six inches in diameter) between it and the mirror. The globe, being filled with a colored solution, acted as a light filter and condenser; by changing the solution the same preparation could be studied with several different kinds of light. Many details were observed in this way which are hardly visible with white cloud illumination.

II. Spindle Formation in Agave.

First Mitosis.

The species studied is *Agave americana* L. The material was fixed in the field; the best results were obtained with Flemming's strong mixture. Flemming's triple stain was used almost exclusively.

The following account relates to the first division of the pollen-mother-cells. The cytoplasm of the resting cell presents the appearance of a network with fairly regular meshes. The fibres of the network are covered to a greater or lesser extent by a deposit of granular substance which tends to stain somewhat more deeply with orange G than the substance of the fibres. The network appears practically alike in all parts of the cell.

1. PROGENETIC STAGE.

During the formation of the dense chromatin thread and the separation of the chromosomes the meshes of the cytoplasm become radially elongated, until they present the appearance seen in fig. 1. The meshes immediately surrounding the nucleus are somewhat smaller and more regular than the others. Just outside these is a layer of somewhat denser and more granular cytoplasm, while the peripheral layer consists of greatly elongated meshes, which produce at first glance the impression of free fibres. The fibres are not free, however, but are constituent parts of a network; it is interesting to observe, nevertheless, that a large proportion of these fibres run more or less in a straight line directly to the nucleus. The layer of cytoplasm, one mesh in thickness, immediately in contact with the nuclear wall, is of quite even width (see fig. 1) and gives rise to the genetic or spindle-forming layer; it will therefore be called the *progenetic layer*, and may be defined as that layer of the cytoplasm from which the genetic layer immediately arises. The stage in which this layer takes on its characteristic condition will be called the *progenetic stage*, and may be defined as the stage immediately preceding the genetic stage, and in which preparation for the latter is going on in the cytoplasm.

In fig. 2 the progenetic layer may be seen undergoing a characteristic change: at one end of the nucleus (the upper end in the figure) it retains its original form, on the sides it is growing narrower, its radial fibres are becoming less distinct, and granules are making their appearance

along its periphery; these begin to stain violet with the Flemming triple stain. At the same time a few delicate fibres are seen along its periphery, which resemble the granules in their manner of staining; they seem, in fact, to be composed largely of granules of the same sort as those just described.

2. Genetic Stage.

The granules in the periphery of the layer increase in number and size and stain deeper and deeper violet, as is seen in figs. 3 and 4. Eventually they become densely crowded, and apparently fuse to form an unbroken line which stains deep violet. Careful focussing shows that this line represents sooner or later a continuous membrane which forms a complete and unbroken investment of the spindle during its formation, and which will therefore be called the *spindle wall*. The spindle wall is very often, at least in the early stages, not in contact with the nuclear wall, but in many cases it coalesces at one or more places with the nuclear wall. This may happen even in the earliest stages, as is shown in fig. 2.

During the formation of the spindle wall there are gradually developed, between it and the nuclear wall, radial fibres such as are seen in fig. 4. These fibres are at first very delicate, stain faintly orange, and contain irregular scattered granules, which tend to stain violet. As the formation of the spindle wall progresses, the granules increase in number and size, become crowded, and finally seem to fuse to produce continuous violet fibres. Thereupon the fibres lose their granular appearance, become thicker, straighter, and stain more and more violet, as is shown in figs. 5–8. Inasmuch as these fibres subsequently form the spindle, they will be called the *genetic fibres*: the layer containing them, i. e., the space between the nuclear and spindle wall, will be called the *genetic layer*, and it is suggested that this term be used generally to designate the layer containing the genetic fibres, whether that layer be bounded by a definite wall or not; the stage which is

characterized by the presence of the genetic fibres will be called the *genetic stage*, and may be defined as lasting from the first appearance of the genetic fibres to the time when the cones are fully developed, and the nuclear wall has disappeared. As in many cases these two processes do not occur simultaneously, it should be understood that the genetic stage is not ended until both are completed.

The genetic fibres are attached to both the nuclear wall and the spindle wall; the point of insertion on the latter is marked by a granular enlargement (see figs. 4 to 10). They may be derived in part from the original radial fibres of the cytoplasmic layer abutting on the nucleus, but to a great extent they appear to be new formations, since they are much more numerous than the original radial fibres. The fact that the nucleolus disappears about this time is in accord with Strasburger's assumption that its substance is used to furnish material for the kinoplasmic fibres (*cf.* Strasburger, 1900, page 124, *ff.*; 1897, page 378, *ff.*; 1895).

During the development of the genetic fibres, the genetic layer gradually widens, and the radial arrangement of the cytoplasm becomes less pronounced (figs. 3 to 7). In the meantime, the irregular fibres lying outside the spindle wall have undergone a change similar to the genetic fibres inside the wall. The former will be called the *exterior fibres* to distinguish them from the genetic fibres. The granules in the exterior fibres become crowded, and fuse just as in the genetic fibres; the fibres become smoother, straighter, thicker, and less granular in appearance, staining deeper and deeper violet. At the same time, they gradually take on a more radial arrangement, and grow longer, until they finally present the appearance seen in fig. 7. The stages in this process are clearly seen in figs. 2 to 7. These fibres appear to be the result of a transformation of the fibres of the cytoplasmic network, the process of transformation proceeding from the spindle wall outward.

It is in connection with certain of these exterior fibres that the cones are formed. It is usually where the more conspicuous exterior fibres (those which extend to the

limiting membrane of the cytoplasm) are attached to the spindle wall that the latter begins to form conical projections such as are seen in figs. 7 and 8. Many very conspicuous fibres seem to be in no way connected with the formation of cones; around these fibres, however, other fibres frequently group themselves to form fibre cones; but this is not followed by the development of conical projections of the spindle wall.

The width of the genetic layer at this time varies from that shown in fig. 7 to a width not exceeding that in the early stage, shown in fig. 3.

The subsequent development of the cones is shown in figs. 8 to 10; the larger ones often extend completely to the limiting membrane of the cytoplasm, and the smaller ones are usually connected with it by a conspicuous fibre.

During this period, the exterior fibres diminish in number, lose their power to stain violet, and begin to stain slightly orange. They are apparently retransformed into cytoplasmic reticulum. They have usually ceased to be recognizable (or at least the majority have) at the beginning of the disappearance of the nuclear wall.

During the development of the cones the genetic fibres contained in them undergo a certain amount of rearrangement, tending to make them converge more or less toward the apex of the cone. At the same time, they lengthen to keep pace with the elongation of the cone, and the granular enlargement at the point of their insertion on the spindle wall gradually disappears as though its substance were being drawn into the fibre to provide material for its elongation.

The pollen-mother-cells of *Agave* are very favorable material for the study of the disintegration and disappearance of the nuclear wall, and the penetration of the genetic fibres into the nuclear cavity. The nuclear wall first becomes granular, and just before it begins to disappear consists of numerous small granules between which a delicate membrane is visible. Thereupon the membrane presents the appearance of being slowly dissolved, growing thinner and

at the same time losing its capacity for staining violet. After it has entirely disappeared, the granules remain intact for some time, and then disappear in turn. The dissolution of the wall begins beneath one of the cones; the wall then breaks down under the others, and the process slowly extends from these points until the wall has completely disappeared (see figs. 10 and 11).

The development of linin fibres keeps pace with that of the genetic fibres, and at the disappearance of the nuclear wall these fibres are quite numerous (see fig. 10).

Usually the spindle wall begins about this time to fray out more or less into free fibres and lose its membranous character. This disintegration of the spindle wall occurs sometimes earlier, sometimes later, and is decidedly variable as regards the time of its commencement.

3. STELLAR STAGE.

With the disappearance of the nuclear wall, and the complete development of the cones, the genetic stage may be considered to end. In the next stage the spindle has a star-shaped appearance, due to the irregular position of the cones; this stage will therefore be called the *stellar stage;* it may be defined as lasting from the end of the genetic stage to the time when the cones are separated into two groups and the fibres begin to take on a parallel arrangement. From this time on the spindles resemble fasces, and this stage will accordingly be called the *fascicular stage;* it may be considered to end with the fusion of the cones to form the bipolar spindle, or, in cases where this does not occur, with the arrival of the daughter chromosomes at the poles.

In *Agave* the beginning of the stellar stage is marked (in the pollen-mother-cells) by the penetration of the genetic fibres into the nuclear cavity (figs. 10 and 11), where they soon become mingled with the linin fibres. The number of poles is usually, at this time, from three to eight.

The chromosomes retain their original arrangement for some time after the disappearance of the nuclear wall (fig. 12),

but gradually become displaced. The fibres increase in number and soon begin to attach themselves here and there to the chromosomes, at first singly, and later in bundles; the latter condition is shown in fig. 13. The fibres now begin to take on a parallel arrangement, and the cones separate into two groups marking the beginning of the fascicular stage. It may be remarked, in passing, that the condition shown in fig. 12, where two of the cones seem to dominate the rest, and the fibres stretched between them tend to be parallel, is but temporary.

4. Fascicular Stage.

The fascicular stage is characterized by the increase in number of the cones, as is clearly seen in fig. 15. This is the result of a rearrangement of the fibres, whereby each cone becomes broken up into several. In the meantime the arrangement of the fibres becomes more parallel, and the chromosomes collect in the region of the equator (figs. 14 and 15); at the same time the mantle fibres begin to develop.

5. Bipolar Stage.

The transition to the bipolar stage is easy to follow in the preparations; the cones gradually fuse until the stage shown in fig. 16 is reached. This, as will be seen in the figure, is not strictly bipolar, but the ends of the spindle are drawn out into two or more long points which often extend to the limiting membrane. In many spindles the fusion is complete at one or both of the poles, so as to form a sharp point; but a large proportion of them remains even during the anaphase, in the condition shown in fig. 16.

During the stellar and fascicular stages the cytoplasm loses almost all trace of its radial arrangement; there is a layer of less dense cytoplasm around the spindle (figs. 10–16).

The completed spindle presents in surface view the appearance shown in fig. 16; there is little or no trace of connecting fibres; in median optical section, however,

these may be seen, and in cross sections they appear as a sort of central core of the spindle (fig. 30). After the chromosomes leave the equator this central core of connecting fibres stands out prominently.

As will be seen in fig. 30, these fibres are connected with each other by small delicate fibres, so as to form a sort of network. It will be remembered that Belajeff (1894) described the spindle of *Larix* as a network, but his description refers to its appearance in longitudinal section. The appearance of the spindle of *Agave* in longitudinal section would not justify his description.

Second Mitosis.

1. INTRODUCTION.

The cell-plate formation following the first mitosis presents very interesting features which will be fully described in another paper. The migration of the chromosomes to the poles reveals the presence of numerous connecting fibres forming a sort of central core of the spindle. These fibres remain for some time closely compacted and then begin to spread apart until they finally extend completely across the cell. In the meantime the cell-plate has commenced to form, and as it nears completion, the connecting fibres begin to disappear. Those in the center are the first to go, leaving a peripheral shell of fibres of great regularity and distinctness, which persists for some time. Finally, this in turn disappears, leaving a completed cell-plate extending from wall to wall.

Coincident with the disappearance of the fibres is the reappearance of the cytoplasm, which begins in the center and extends toward the periphery of the space previously occupied by the spindle fibres. After the completion of the cell-plate it increases rapidly in thickness, especially where it joins the cell-wall, until the condition shown in fig. 17 is reached.

The daughter nucleus has in the meantime grown to its full size, and presents the appearance shown in fig. 17.

2. PROGENETIC STAGE.

The region occupied by the spindle remains distinctly marked for some time owing to the fact that the cytoplasm contained within it is less granular and less dense than the rest (figs. 17–22). Throughout the entire cell the cytoplasm shows a marked tendency to a radial arrangement, being denser in the neighborhood of the nucleus (fig. 17).

The first indication of spindle formation is the appearance of granules (which stain violet) in the layer of cytoplasm immediately adjoining the nucleus. They are imbedded in the strands or fibres of the cytoplasmic network and are first seen close to the nuclear wall, from which they gradually extend outward into the cytoplasm. They are at first irregularly scattered, but soon become more numerous and more closely crowded together until finally they appear to fuse together to form continuous fibres which are at first granular and irregular in outline, but gradually become regular and smooth in appearance (fig. 18).

3. GENETIC STAGE.

The fibres so formed are attached to the nuclear wall and their formation always begins at the wall and proceeds outward towards the periphery of the cell. The fibres at first are wavy and irregular and follow the outlines of the cytoplasmic meshes, but gradually straighten out and come to lie in a radial direction (fig. 19). Since these fibres go to form the spindle, they will be called the genetic fibres. The genetic fibres gradually increase (figs. 20 and 21) in number and size—some being much larger and more prominent than others—until the stage shown in fig. 22 (upper half) is reached. They then begin to assemble in groups, as shown in the lower half of the same figure, in which various stages of the process are shown. So far as can be judged from their appearance, the groups are formed simply by a change in position of the fibres, whereby their free ends gradually come together at certain places and finally fuse to form cones. The location of the cones is variable;

the number is usually not less than four or more than ten. Not all the fibres are used in the formation of cones, since some remain, like the exterior fibres of the first division, radiating out into the cytoplasm after the cones are fully formed (fig. 23).

The disappearance of the nuclear wall begins, as is usually the case, under one of the cones, and gradually continues under first one and then another of the remaining cones until complete. The genetic fibres penetrate rapidly into the interior of the nucleus and become attached to the linin network, which increases rapidly during cone formation (fig. 23, lower half).

The details of the process just described are extremely variable. Sometimes the formation of the genetic fibres is greatly delayed, and scarcely anything can be seen of such fibres until just before the nuclear wall breaks down; then, however, they appear to be formed very rapidly. In such cases the cones do not usually reach the size shown in fig. 23.

During the genetic stage the nucleolus disappears completely while the linin increases in amount (fig. 23).

From the figures it will be seen that while in general the process of spindle formation goes on simultaneously in both daughter cells, this does not quite hold true of such stages as are shown in figs. 22 and 23. This is probably to be accounted for by the fact that these stages are passed through with extreme rapidity. These remarks apply particularly to the assemblage of the fibres into cones and the breaking down of the nuclear wall. The stage shown in fig. 23, upper half, is not passed through quite so rapidly, and it is more common to find both daughter cells in this stage at the same time.

4. STELLAR STAGE.

The stellar stage is variable in length and presents considerable diversity as regards the form of the spindle and the number of cones composing it. Frequently the cones

are prominent and well defined, and may reach nearly to the limiting membrane (figs. 24 to 26), while in other cases they are merely small, ill-defined elevations. Free fibres, which radiate out into the cytoplasm, persist for a time, but gradually disappear. The cones are generally unequal, some having larger, longer, more numerous and more prominent fibres than others.

The chromosomes preserve for a time their original arrangement (fig. 25), but gradually become displaced and irregularly scattered (figs. 25 and 26). The fibres in the meantime combine to form more or less definite strands which are at first irregular and loosely aggregated, but which gradually become more compact and regular in appearance. Gradually these strands or groups of fibres become attached to the chromosomes; the attachment is at first partial, involving only a few of the fibres of the strand, but gradually extends to the other fibres of the group (fig. 26).

5. FASCICULAR STAGE.

While the attachment of the fibrous strands to the chromosomes is taking place the arrangement of the strands themselves begins to undergo a change whereby they tend to place themselves parallel to one another (fig. 26); at the same time, certain cones lying at the ends of the incipient spindle become increasingly prominent, and seem to serve as places of assemblage for the other cones which appear to gather around them, thus finally forming two distinct groups which lie at the opposite ends of the fascicle (fig. 28). In the meantime, the strands of fibres become more and more compact, and form well defined, thick fibres (fig. 27).

The cones in each group gradually approach each other more closely, and at the same time the number of cones increases (fig. 28); the fusion of the cones is gradual, and frequently takes place sooner at one end of the spindle than at the other (fig. 28). The assemblage of chromosomes in the equatorial plate does not take place until the fusion is well advanced.

6. BIPOLAR STAGE.

Strictly speaking, there is not, as a rule, a bipolar stage, since the fusion does not become complete but remains in the stage shown in fig. 29. It is not uncommon to find the fusion complete at one end but not at the other, and in some cases the spindle may become truly bipolar.

GENERAL CONSIDERATIONS.

The spindle formation of the first mitosis of *Agave*, as set forth above, differs markedly from anything hitherto described. The formation of a spindle wall which completely encloses the spindle during the genetic stage is unique; and the origin and development of the genetic fibres is peculiar.

The spindle wall would seem to be a structure of great physiological importance, comparable in its functions with the nuclear wall and the limiting layer of the cytoplasm. Its special function appears to be to segregate the important process of spindle formation, thus carrying the division of labor in the cell to a point otherwise impossible. Its presence therefore indicates a high degree of cell organization.

In appearance and mode of origin the spindle wall resembles the nuclear wall; its close relation to the nucleus is shown by its origin and development; it persists, however, after the nuclear wall has disappeared, and may then perhaps assume some of its functions. It is either in actual contact with the nuclear wall during the genetic stage or is constantly connected with it by the genetic fibres; on the other hand, it early forms connections with the limiting membrane by means of the exterior fibres. The relations existing between the kinoplasm and the limiting membrane have recently been emphasized by Strasburger (1900, p. 144 *et seq.*), who calls attention to the fact that in many forms the kinoplasmic fibres can be traced to this membrane and that the spindle is in many cases inserted on it. *Agave* offers direct confirmation of these statements.

It is possible that there may be some homology between

the spindle wall and the structures, more or less membranous, which form at varying distances from the nuclear wall in the pollen-mother-cells of *Lilium* (Mottier, 1897), *Cobæa* (Lawson, 1898), and *Lavatera* (Byxbee, 1900). These structures consist at first of a rather loose weft of fibres (which stain violet with the Flemming stain); later the fibrous weft becomes condensed and compressed so as to form in many cases a thick continuous membrane which stains violet; other cases may usually be found in the same anther, in which the membrane is not continuous or frays out in places into a fibrous weft. These structures stand in no such intimate relation with the nucleus and the spindle as the spindle wall of *Agave*, and seem in comparison with it irregular and ill defined. That they may have some phylogenetic connection with the spindle wall is not improbable; if so, they are probably to be regarded as rudimentary structures.[1]

On the other hand, it seems quite certain that many of the important resemblances between animal and plant cells are to be regarded not as homologous but as analogous, and the same is probably true of resemblances in cell structure between different groups of plants. We must therefore exercise considerable caution in assuming homologies between cell structures.

The development of the genetic fibres is peculiar in that there is no weft stage such as is characteristic of most forms of higher plants hitherto studied; on the contrary, the fibres are radial from the beginning. This may very likely be connected with the presence of the spindle wall. Numerous observations incline me to believe that the weft stage is caused by the expansion of the nucleus, which at first causes a tangential elongation of the meshes immediately surrounding the nucleus; as the expansion continues the meshes are broken and form free fibres. It is easy to see how the turgidity of the genetic layer (contained within the

[1] An interesting case is that described by Eisen (1900) for *Batrachoseps*, where a "false nuclear wall" is formed around the daughter chromosomes: within this the true nuclear wall is formed.

spindle wall) could hinder or altogether prevent the expansion of the nucleus and so eliminate the weft stage. The osmotic pressure of the genetic layer, if equal to or slightly greater than that of the nucleus, would bring about this result.

The exterior fibres do not seem to be essentially different from the genetic fibres save that, being outside the spindle wall, most of them take no direct part in the formation of the spindle. Both kinds of fibres seem to be similar to the radial kinoplasmic fibres of *Equisetum*. Some of the exterior fibres form fibre cones in much the same way as in *Equisetum*, but these structures are, in the case of the first mitosis of *Agave*, merely temporary.

The formation of the spindle cones is peculiar on account of the part played by the exterior fibres. According to Belajeff (1894), the spindle cones of *Larix* are formed in connection with certain fibres which extend from the genetic layer to the cell wall. In this respect the process resembles that which occurs in the first mitosis of *Agave*, but the fibres do not group themselves to form the spindle cones in the manner described by Belajeff.

While the method of spindle formation in the first mitosis stands unique, that of the second mitosis recalls the origin of the spindle in the spore-mother-cells of *Equisetum* (Osterhout, 1897), with which it agrees in most of its principal features, *e. g.*, in the origin of the genetic fibres in contact with the nuclear wall, their radial arrangement and outward growth into the cytoplasm, their assemblage in groups accompanied by a fusion of their free ends to form cones, and, finally, the fusion of these cones to form the spindle. In *Equisetum*, however, there is at first a modified weft stage of which no trace is seen in *Agave*. This agreement with *Equisetum* is the more surprising since the relationship between the two plants is so remote, while the spindle formation of the immediately preceding mitosis of *Agave* presents such profound differences.

Investigations on spindle formation, though as yet but few in number, have revealed great diversity, especially in

plants, where almost every careful investigation has discovered a new type. Widely divergent types have been shown to occur in different tissues in the same individual (Nemec, 1898; Strasburger, 1900, p. 119, also, p. 112, *ff.*) or even in the same tissue (Davis, 1901; Hertwig, 1898). In view of this we may conclude that the process of spindle formation is a plastic one, easily modified in the course of phylogeny and even of ontogeny, and that its study deserves to be seriously taken up as a matter of vital importance to an understanding of cell mechanics.

SUMMARY.

The first spindle formation of the pollen-mother-cells of *Agave americana* L. presents the following features of especial interest.

1. During the early stages of the spindle it is enclosed in a special membrane (of cytoplasmic origin) which forms a complete investment around it, and is called the *spindle wall;* the functions of this membrane appear to be comparable to those of the nuclear wall and the limiting membrane of the cytoplasm; it appears to be a unique structure.

2. Unlike most cases hitherto described, no weft stage is present; this is probably due to the presence of the spindle wall, which prevents the expansion of the nucleus, and consequently the formation of the weft.

3. The genetic (*i. e.*, spindle-forming) fibres are radial from the beginning, and are attached to both the nuclear and the spindle wall.

The second spindle formation differs *in toto* from the first and may be summarized as follows: The genetic (spindle forming) fibres form in close contact with the nuclear wall, take on a radial arrangement, extend outward into the cytoplasm, assemble in groups, and form cones which by their fusion give rise to the spindle. The process resembles in general the spindle formation of the spore-mother-cells of *Equisetum*.

It is proposed that the stages of spindle formation be characterized as

(1) Progenetic
(2) Genetic
(3) Stellar
(4) Fascicular
(5) Bipolar.

The *progenetic stage* may be defined as the stage preceding the genetic, and in which preparation for the genetic stage goes on in the cytoplasm; the region of the cytoplasm in which this takes place and from which the genetic layer arises may be called the *progenetic layer*.

The *genetic stage* lasts from the first appearance of the *genetic fibres* (*i. e.*, the spindle-forming fibres) to the complete development of the spindle cones and the disappearance of the nuclear wall. When these processes do not take place simultaneously, it is to be understood that the genetic stage does not end until both are completed. The layer containing the genetic fibres is called the *genetic layer*.

The *stellar stage* lasts from the end of the genetic stage to the separation of the cones into two (more or less opposite) groups, and the commencement of the parallel arrangement of the fibres.

The *fascicular stage* lasts from the end of the stellar stage to the complete fusion of the cones (to form the bipolar spindle), or, where this does not occur, to the arrival of the daughter chromosomes at the poles.

The *bipolar stage* lasts from the end of the fascicular stage to the complete disappearance of the poles.

LITERATURE CITED.

1894. BELAJEFF, W. Zur Kenntniss der Karyokinese bei den Pflanzen. *Flora*, Bd. LXXIX, p. 430.

1900. BYXBEE, E. S. The Development of the Karyokinetic Spindle in Lavatera. *Proc. Cal. Acad. Sci.*, 3d Ser. (Bot.), Vol. II, p. 63.

1901. DAVIS, B. M. Nuclear Studies on Pellia. *Annals of Botany*, Vol. XV, p. 147.

1897. EISEN, G. Notes on Fixation, Stains, the Alcohol Method, etc. *Zeitschr. f. wiss. Mikr.*, Bd. XIV, p. 195.

1900. ——The Spermatogenesis of Batrachoseps. *Journ. Morph.*, Vol. XVII, p. 1.

1898. HERTWIG, R. Ueber Kerntheilung, Richtungskörperbildung und Befruchtung von Actinosphærium Eichornii. *Abh. d. k. bayr. Akad. d. wiss.* II Cl., Bd. XIX, p. 636.

1898. LAWSON, A. A. Some Observations on the Development of the Karyokinetic Spindle in the Pollen-Mother-Cells of Cobæa scandens Cav. *Proc. Cal. Acad. Sci.*, 3d Ser. (Bot.), Vol. I, p. 169.

1900. ——Origin of the Cones of the Multipolar Spindle in Gladiolus. *Bot. Gazette*, Vol. XXX, p. 145.

1897. MOTTIER, D. M. Beiträge zur Kenntniss der Kerntheilung in den Pollenmutterzellen einiger Dicotylen und Monocotylen. *Jahrb. f. wiss. Bot.*, Bd. XXX, p. 169.

1898. NEMEC, B. Ueber die Ausbildung der achromatischen Kerntheilungsfigur, etc. *Bot. Centralblatt*, Bd. LXXIV, p. 1.

1897. OSTERHOUT, W. J. V. Ueber Entstehung der karyokinetischen Spindel bei Equisetum. *Jahrb. f. wiss. Bot.*, Bd. XXX, p. 159.

1900. ——Befruchtung bei Batrachospermum. *Flora*, Bd. LXXXVII, p. 109.

1895. STRASBURGER, E. Karyokinetische Probleme. *Jahrb. f. wiss. Bot.*, Bd. XXVIII, p. 151.

1897. ——Ueber Cytoplasmastructuren. Kern- und Zelltheilung. *Jahrb. f. wiss. Bot.*, Bd. XXX, p. 375.

1900. ——Ueber Reductionstheilung, Spindelbildung, Centrosomen und Cilienbildner im Pflanzenreich. Jena.

1899. WILLIAMS, C. L. The Origin of the Karyokinetic Spindle in Passiflora cœrulea Linn. *Proc. Cal. Acad. Sci.*, 3d Ser. (Bot.), Vol. I, p. 189.

EXPLANATION OF PLATE XXV.

The material was fixed in the field with Flemming's strong mixture; sections were cut 1–5 μ thick and stained with Flemming's triple stain. Figures were drawn with the aid of the Abbé camera lucida, Zeiss' Apoc. Hom. Im. Obj. 2.00 mm., Comp. Oc. 8. All pertain to the division of the pollen-mother-cells of *Agave americana* L.

The lithographic plates reproduce the details of colour and form of the preparations with great fidelity; the differentiation of the kinoplasmic elements is in no way exaggerated.

Fig. 1. Pollen-mother-cell before the beginning of spindle formation. The cytoplasm shows the characteristic radial arrangement, the inner portion being denser.

Fig. 2. Progenetic layer in process of transition to the genetic (spindle forming) stage. It shows violet granules along its periphery, and outside this a few delicate fibres which later develop into the exterior fibres.

Fig. 3. The periphery of the layer more violet; exterior fibres more developed.

Fig. 4. Periphery of the layer shows violet granules more closely crowded together; radial genetic fibres appear within the layer; exterior fibres still more developed; *beginning of genetic stage*.

Fig. 5. Spindle wall definitely formed; genetic and exterior fibres still more developed.

Fig. 6. Genetic layer widening; exterior fibres begin to form fibre cones.

Fig. 7. Beginning of cone formation.

Fig. 8. Further development of the spindle cones.

EXPLANATION OF PLATE XXVI.

Fig. 9. The spindle cones completely developed; the exterior fibres disappearing.

Figs. 10 and 11. Disappearance of the nuclear wall; *beginning of stellar stage.*

Fig. 12. The nuclear wall has completely disappeared, but the chromosomes retain their original arrangement. Two of the cones seem to dominate the rest, but this is only a temporary arrangement.

Fig. 13. Beginning of *fascicular stage;* the fibres are assuming a parallel arrangement; the cones are separated into two groups; fibres are becoming attached to chromosomes in bundles.

Fig. 14. The parallel arrangement of the fibres and their attachment to chromosomes is more pronounced.

Fig. 15. The cones have increased in number; the chromosomes are assembled in the equatorial region.

Fig. 16. Completed spindle; not strictly bipolar, since each end is drawn out into two or more points.

EXPLANATION OF PLATE XXVII.

Fig. 17. Progenetic stage of second mitosis. The cytoplasm shows a radial arrangement; in the region previously occupied by the spindle fibres of the first mitosis the cytoplasm is less dense. Granules which stain violet begin to make their appearance in strands of cytoplasm nearest the nucleus.

Fig. 18. Genetic stage. The violet granules gradually increase in number, become crowded, and fuse to form the genetic fibres.

Fig. 19. The genetic fibres increase in number and size.

Figs. 20 and 21. The genetic fibres increase and assume a more regular arrangement. The lower half of the figure shows in both cases a slightly more advanced stage than the upper.

Fig. 22. The upper half shows the genetic fibres fully formed; in the lower half they are assembling in groups to form the spindle cones; various stages in this process are shown.

Fig. 23. The upper half shows the cones fully formed: the lower half shows the breaking down of the nuclear wall and the penetration of the genetic fibres into the nucleus, marking the commencement of the stellar stage.

Fig. 24. The nuclear wall has completely disappeared; the chromosomes preserve their original arrangement.

EXPLANATION OF PLATE XXVIII.

Fig. 25. In the upper half of the figure the chromosomes still preserve their original arrangement; in the lower half they have already become somewhat disarranged. The genetic fibres tend to collect into strands which have not yet attached themselves to the chromosomes.

Fig. 26. The fibrous strands now show a more or less complete attachment to the chromosomes and a tendency to assume a parallel arrangement.

Fig. 27. The strands have become compact fibres and are more nearly parallel.

Fig. 28. Transition to bipolar stage. In the lower half of the figure the chromosomes are assembling in the equatorial region.

Fig. 29. Completed spindle; not strictly bipolar, since the fusion of cones is not complete.

Fig. 30. Cross-section of spindle of first mitosis near the equatorial region, showing the central core of connecting fibres interconnected by delicate threads.

EXPLANATION OF PLATE XXVII.

Fig. 17. Progenetic stage of second mitosis. The cytoplasm shows a radial arrangement; in the region previously occupied by the spindle fibres of the first mitosis the cytoplasm is less dense. Granules which stain violet begin to make their appearance in strands of cytoplasm nearest the nucleus.

Fig. 18. Genetic stage. The violet granules gradually increase in number, become crowded, and fuse to form the genetic fibres.

Fig. 19. The genetic fibres increase in number and size.

Figs. 20 and 21. The genetic fibres increase and assume a more regular arrangement. The lower half of the figure shows in both cases a slightly more advanced stage than the upper.

Fig. 22. The upper half shows the genetic fibres fully formed; in the lower half they are assembling in groups to form the spindle cones: various stages in this process are shown.

Fig. 23. The upper half shows the cones fully formed: the lower half shows the breaking down of the nuclear wall and the penetration of the genetic fibres into the nucleus, marking the commencement of the stellar stage.

Fig. 24. The nuclear wall has completely disappeared; the chromosomes preserve their original arrangement.

lobed with mucronate or obtuse lobes, 2–6 cm. wide at base, 2–5 cm. long. Flowers chiefly near the base, on long peduncles surpassing the leaves, often upwardly curving, 5–8 cm. long; bracts similar to the upper leaves, close under the flower, 1 cm. wide, 1.5 cm. long. Sepals ovate-oblong, unequal in breadth but of the same length, hairy with appressed hairs except on the membranous margins of the inner ones, mucronate, 14 mm. long, the broadest 10 mm. wide. Corolla yellowish white, hairy on the angles and at their tips, 4 cm. long. Stamens with narrow sagittate anthers, 7 mm. long; filaments hairy below, attached to the corolla for half their length. Style as long as the stamens (2.5 cm.), with ovate-lanceolate stigmas, 3 mm. long, 1 mm. wide.

This beautiful species comes nearest to *C. villosus* Gray, from which it differs in the bracts, peduncles, and stamens, also in the broader leaves and longer peduncles. The pubescence is less velvety. It is also near *C. tomentellus* Greene, from which it differs in almost the same organs.

Collected at Millwood by the author, July, 1893 and 1899, also near Converse Basin. The plant is named in honor of Mr. S. L. Berry.

6. **Castilleia brooksii**, sp. nov.

Perennial, branching from the base, but with a few short branches above, 3 dm. high, glandular, viscid throughout and with a pubescence of uneven, silvery, weak, jointed hairs. Leaves sessile, linear-oblong, entire to three-lobed, about 2 cm. long, less than 5 mm. wide; lobed leaves generally subtending the branches, the divided portion half the length. Branchlets terminated by short, compact spikes, with subsessile flowers; bracts variously and unevenly lobed, with the tips colored. Calyx a little longer than the corolla tube, obliquely gibbous at base, equally cleft before and behind, with divisions shorter than the tube, each two-cleft, with unequal, triangular-subulate, one-nerved, obtuse divisions, 3 mm. long. Corolla 2.5 cm. long; galea longer than the tube, straight at first but later curving outwards, having three blunt teeth at apex, the middle one smallest; lower lip truncate, 3 mm. long, 2 mm. wide, the sharply acute teeth incurved, folds noticeable. Stamens exserted, with filaments smooth and anthers narrowly linear, with unequal cells. Ovary glabrous, obliquely acuminate; stigma clavate, exserted from the top of the galea.

This comes under the group to which belong *C. parviflora* Bongard, and *C. miniata* Douglas, with neither of which it agrees. The flowers are yellowish red but probably variable in color, as in most species of this genus.

Collected by the author on the trail up Bubbs Creek, early in July, 1899, and named in honor of Mr. Benjamin Brooks.

7. Castilleia disticha, sp. nov.

Perennial, erect, 6 dm. high, branching from the base and also above with generally short, slender, spreading branches; somewhat viscid and with a close, often somewhat scanty, cinereous pubescence mixed with longer, jointed hairs. Leaves linear, acute or obtuse, the lower 4 cm. long, 3 mm. wide, diminishing upwards, sessile by a truncate or subauriculate base, distinctly three-veined, the middle vein most conspicuous; margin entire or undulate-crisped. Inflorescence spicate, elongating in fruit, especially on the main stem, 1–2 dm. long; flowers after anthesis distichous, becoming more or less remote, sessile or almost so, with the capsule appressed to the stem, the calyx and corolla persisting and spreading; bracts foliaceous, the upper ones, only, colored, variously toothed, with the middle tooth longest, equalling or shorter than the corolla. Calyx slightly surpassing the corolla tube, about equally cleft before and behind for half the length; each division tipped with red, two-cleft, with triangular, subulate, unequal lobes, 2–3 mm. long, three-nerved, thin in texture, somewhat gibbous, but not broadest at base. Corolla red, 3 cm. long, with galea as long as the tube, truncate or emarginate at apex; lower lip three-toothed, the middle tooth much smaller than the lateral, separated by a broad sinus, thin, not callous. Stamens exserted, filaments glabrous; anthers narrow, with unequal cells, more than 2 mm. long. Stigma exserted from the summit of the galea, capitate. Capsule obliquely oblong-ovate, 8–10 mm. long, chartaceous; seeds elliptical to orbicular, light brown, invested with a membranous, foveolate outer coat.

This is more closely allied to *C. minor* Gray and *C. stenantha* Gray than to any of the perennial species. Its sessile or almost sessile flowers, more brightly colored and differently shaped, together with the different habit of growth, mark it as distinct.

The type was collected by the author at Converse Basin, on the trail to the south fork of Kings River, July, 1899. The species is also found at Millwood, where it was collected the same year by the author, and by Mr. T. S. Brandegee, July 19, 1892.

8. Castilleia nana, sp. nov.

Low, 3–6 cm. high, with several stems from a woody caudex, which is thickly clothed with the dead stems of former seasons; somewhat cinereous and viscid, the pubescence of the inflorescence of longer, jointed, arachnoid hairs. Leaves simple and linear or three- to five-divided, with narrow, linear, acuminate lobes which are shorter or longer than the undivided portion, together 1–2 cm. long. Flowers in heads terminating the stems; bracts broad, similar to the upper leaves, embracing the sessile flowers and more than twice as long; the division terete from the involute margins, once and a half to twice as long as the lower, undivided part, which is 4 mm. wide

and 5 mm. long. Calyx glandular, subequally cleft before and behind, with the divisions twice as long as the tube, surpassing the corolla, each with two narrow, linear lobes 5-6 mm. long, extending below the throat of the inconspicuous corolla. Corolla 7 mm. long, the lower lip longer than the galea, the three lobes thin, ovate-orbicular, obtuse, 2 mm. wide, somewhat saccate below; galea with the middle portion of thicker texture than the membranous sides, three-toothed at apex. Stamens with the upper pair of anthers exserted, the lower included, each distinctly two-celled, clothed with a few long hairs at base. Pistil capitate, .5 mm. in diameter, exserted; ovary lanceolate. Fruit and seeds unknown as the plants were too young.

This is an alpine species found only above timber line. It is related to the group formerly included under *Orthocarpus*, section Castilleioides, but differs from all other species in the peculiar corolla, it being the only species described with the galea shorter than the lower lip.

Collected by the author on Harrison's Pass, above East Lake, July 9, 1899.

There are either one or two other low, alpine species in the same region which seem to be related to *C. pallida* Kunth, but the material is too scanty for satisfactory determination.

9. **Mimulus bioletti**, sp. nov.

Annual, branching diffusely, chiefly from the base, about 2 dm. high, glandular-villous throughout, except the corolla. Leaves near the root spatulate; cauline leaves rhombic obovate to lanceolate, thin, tapering to a broad petiole, or sessile, sparingly serrulate or entire, 2-4 cm. long, 2-15 mm. broad. Flowers axillary, on slender, upward-spreading peduncles, almost as long as the internodes, generally shorter than the subtending leaves. Calyx tubular, 8 mm. long when in flower, 12 mm. in fruit, often purplish-dotted below the middle, plicately carinate-angled, the ribs rugose, rounded; divisions deltoid, with the margins involute, the obtusely pointed apex spreading outwardly. Corolla as long again as the calyx, with ampliate throat and scarcely two-lipped border, crimson, the upper lip with a yellow blotch dotted with crimson in the throat; limb 12-15 mm. across, with divisions rounded, crenulate, or entire. Stamens and style included; the former four in two sets, each united by the anthers, one set longer than the others; anthers ciliate, explanate, one above the other; stigma bilammelar, cuneate in outline. Capsule included in the rigid calyx-tube, obtusely four-ribbed, opening at the sides from the base up; placenta free, except at the top; seeds numerous, minute.

This belongs to section Eumimulus Gray, and is most closely related to *M. palmeri* Gray. It differs chiefly in the larger flowers and different calyx.

Collected in Hetch-Hetchy Valley, Tuolumne County, by Mr. F. T. Bioletti, in July, 1900.

10. **Phacelia stimulans**, sp. nov.

Stems tall, simple from a branched caudex, becoming 5–6 dm. high, erect, sparsely leaved, generally flowering from the middle, viscid-pubescent, and clothed besides with fine, long, stinging hairs. Radical leaves forming a rosulate cluster, simple or with a few lobes at base, ribbed between the hispid veins, elliptical, acuminate, 3 cm. long; petioles very hispid with spreading hairs. Spikes of the panicle simple, the lowest and uppermost geminate, horizontally spreading, somewhat distant, 5–6 cm. long; peduncles very glandular, becoming shorter near the top; pedicels capillary, half as long as the calyx. Divisions of the calyx oblong-spatulate, hispid, net-veined, shorter than the corolla, surpassing the capsule. Corolla tubular, the lobes conniving after anthesis and persistent, held to the calyx by the tangling together of the long, persistent stamens and style. Filaments exserted, conspicuously clothed with long white wool. Capsule ovate-acuminate, hispid; seeds ovate, brown, not glossy, pitted.

This is allied to *P. circinata* Jacq. f., but is entirely unlike any of the described species which were formerly included under that species. On account of the stinging hairs of the stems and leaves it might be confused with *P. nemoralis* Greene; but this has an altogether different habit, pubescence, and inflorescence.

Collected by the author, July, 1899, in Kings River Cañon, not far from the swampy meadow near which campers stop on the way to Bubbs Creek.

11. **Gilia sparsiflora**, sp. nov.

Annual, a foot or so high, branching above, with slender, spreading stems, minutely glandular-pubescent. Leaves few, terete from the infolding of the margins, about an inch long, tipped with a short bristle. Flowers few, terminating the branchlets, two to three in the clusters, sometimes solitary in the upper axils; bracts keeled at base, three-lobed, the middle lobe much larger than the lateral, all subulate-aristate, surpassing the flowers. Calyx membranous between the ribs, clothed with dense, white, cottony wool, the unequal, aristate-subulate divisions as long as the corolla tube. Corolla salver-form, 1 cm. long, white with some purple dots in the funnel-form throat; the divisions elliptical-obtuse, half as long as the tube. Stamens equally inserted, with arrow-shaped anthers, obtuse at apex, exserted from the throat of the corolla. Capsule oblong, 1 cm. long; seeds few, oblique at base, three-sided, generally with rounded angles, developing mucilage and spiracles.

Gilia sparsiflora belongs to the same group as *G. virgata* Steud.

Collected by the author in Kings River Cañon, in July, 1899; and also along Bubbs Creek trail.

12. Cryptanthe vitrea, sp. nov.

Annual, with several stems from a tap-root, 1–2 dm. high, very hispid throughout with white bristly hairs which are pustulate at base. Leaves linear, 1–2 cm. long, 2 mm. wide, strongly nerved. Flowers sessile, in numerous short spikes from almost the lowest axils, on slender peduncles. Calyx 3 mm. long, the sepals conniving to form a tube around the nutlets, the tips free, densely clothed with long stiff bristles, 3 mm. long, and also a fine, white, hispid pubescence. Corolla only about 2 mm. long, with a small limb. Nutlets, only two maturing, ovate, obtuse, almost 2 mm. long and 1 mm. wide at base, very glossy brown mottled, sharply tuberculate, attached to the gynobase for the entire length, the groove closed except at the forks.

The species comes nearest to *C. muriculata* Greene, but the nutlets are broader, the flowers much smaller, and the entire plant so floriferous that in the dried specimen it is almost impossible to distinguish the peduncles.

Collected by the author on Bubbs Creek trail, July 5, 1899.

13. Aster durbrowi, sp. nov.

Stems erect, perennial from creeping root-stocks, disposed to grow in tufts, 1–3 dm. high, sparingly leafy, erect, branching only at the inflorescence, glabrous and green except for some white woolly hairs which are sparse on the lower part, but which make the upper part almost cinereous. Radical leaves on long winged petioles which are dilated and clasping at base, lanceolate, the blades about as long as the petiole, together 5–8 cm. long, 5–10 cm. wide, entire or distantly serrate, ciliate on the margin, the hairs becoming longer towards the base of the petiole and decurrent on the stem; cauline leaves similar but sessile by an auriculate or cordate clasping base, the upper ones broadening and becoming shorter and more pointed, the lower ones narrowing towards the insertion. Heads cymose, the branchlets terminated by one to four middle-sized heads, 1–2 cm. across, on short bracteate pedicels; involucre of six rows of imbricated bracts with green, foliaceous tips, not spreading, the inner ones with purplish acuminate tips, the outer linear, mucronate, glabrous except for the ciliate margins, distinctly one-nerved and chartaceous at base; rays pistillate, reddish purple, 1 cm. long, 1.5 mm. wide, dentate at apex, sparingly ciliate on the lower part; disk flowers purplish; corolla shorter than the pappus, which extends

about to the exserted style and stamens; the tube slightly pubescent. Akenes (immature) hispid with dense, white, upwardly appressed hairs; pappus simple, scabrous.

This is near *A. yosemitanus* Greene (*A. ascendens yosemitanus* Gray), of which it may prove to be a variety. The habit of growth is quite unlike that of the above species, while its larger heads, sparingly leafy stems terminated by few heads, and the auriculate- or cordate-clasping cauline leaves make it appear even more distinct. It is the common aster of the wet meadows at the upper altitudes in this region.

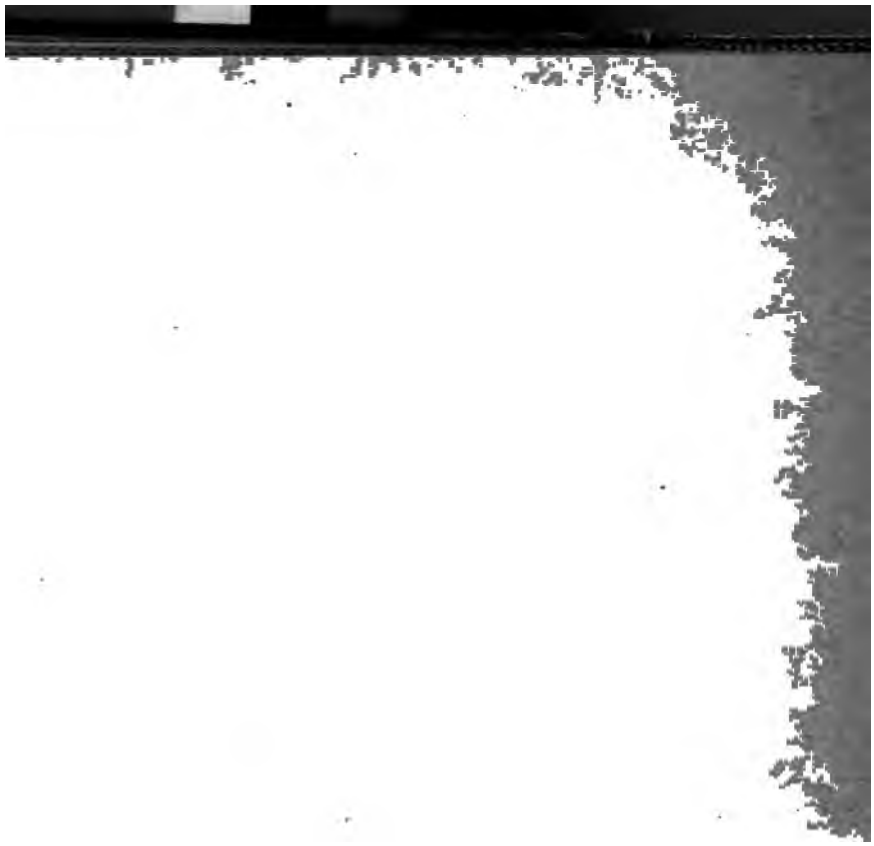
Collected in Horse Corral Meadow, July 11, 1899, and named in honor of Mr. Pierson Durbrow.

14. **Madia villosa**, sp. nov.

Stems simple, erect from an annual root, about 3 dm. high, slender, villous with long, white, soft, spreading hairs, also becoming glandular near the top with black, stipitate glands. Lower leaves generally opposite, upper alternate, linear or the lowest oblanceolate, sessile, entire, or glandular-serrate with distant teeth, revolute, strongly ribbed, villous, with the hairs finely pustulate on the older leaves, obtuse at apex, 4–7 cm. long, 2–5 mm. wide. Heads few, terminating slender peduncles near the top of the stem, the uppermost first in bloom, clothed with few small bractlets; outer bracts of the involucre 8 mm. long, the foliaceous tips equalling that which encloses the akene, linear acuminate, villous and glandular, half as long as the deeply three-lobed rays, these often with a reddish-brown spot at base; inner bracts scarious with short foliaceous tips; disk flowers all sterile, the corollas as long as the abortive akenes, together 8 mm. long, the tube somewhat villous and the lobes clothed at tip with spreading hairs; fertile akenes flattened laterally, semilunate, black and brown mottled, minutely papillate in rows, 1.5 mm. wide, 4 mm. long, glabrous; anthers purple and exserted, giving a purplish color to the disk.

This is nearest to *Madia corymbosa* DC. (*Madaria corymbosa* Greene). It is also close to *M. hispida* Greene, and really seems to be intermediate between the two species.

The type was collected at Converse Basin, July 12, 1899. It was also found on Bubbs Creek trail.



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The Root-tubercles of Bur Clover

(Medicago denticulata Willd.)

and of Some Other Leguminous Plants.

BY

GEORGE JAMES PEIRCE

Associate Professor of Plant Physiology in the Leland Stanford Junior University.

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THE ROOT-TUBERCLES OF BUR CLOVER (MEDICAGO DENTICULATA WILLD.) AND OF SOME OTHER LEGUMINOUS PLANTS.

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I. INTRODUCTION AND METHOD.

SOME time ago, on casually examining some hand-sections of the root-tubercles of Bur Clover (*Medicago denticulata* Willd.), I was struck by the great differences between the cells containing bacteria or bacteroids and those in which there were none. In these sections, the bacteria-containing cells looked so unhealthy, as compared with the cells free from bacteria, as plainly to suggest that the intimate relations of bacteria and leguminous cells were not mutually advantageous, but that the bacteria were parasitic. Thereupon I began a careful microscopic study of the root-tubercles of Bur Clover and other leguminous plants[1] in

[1] Bur Clover was especially favorable for my work because, at most seasons, I could get living plants from out of doors very near the laboratory, and I could grow such material as I needed at other times from seed very quickly in the laboratory. Besides this, however, I have studied *Lupinus micranthus* Dougl. var. *bicolor* Watson, *L. rivularis* Dougl. var. *latifolius*, *Melilotus parviflora* Desf., *Medicago sativa* Linn., *Hoffmannia subpinnata* Torr. and Gray. The points which I wished to determine are essentially the same in all of these, and hence my descriptions, though specifically of Bur Clover, are applicable to the others.

order, if possible, to ascertain the real relations of the tubercle organism to the cells in which it is found. This I believe I have done. The bacteria are parasites, not benefiting but injuring, if not finally killing, the cells in which they occur. Whether the association of these bacteria with a leguminous plant benefits the plant as a whole is another question, answerable not from microscopic examination but solely by experimental cultures. According to Frank (1890, p. 105), the *Leguminosæ* are not all similarly affected by the bacteria. Some are greatly benefited, stimulated to increased growth and other activities, while in others the bacteria are ordinary parasites, not benefiting the host in any way in return for the food derived from it. Without implying whether the results of infection benefit or injure the plant as a whole, one may speak of the roots or cells of leguminous plants being *infected* by tubercle bacteria. I shall later, however, take occasion to discuss whether the presence of the tubercle organism is really beneficial.

The material studied was either fresh, growing out of doors wild or sown in boxes in the laboratory, or alcoholic. The latter was fixed in Flemming's chrom-osmic-acetic mixture, dilute, and after washing for twelve to twenty-four hours in running water was dehydrated and kept in 90 per cent. alcohol. These tubercles, which were of different sizes, ages and conditions, according to the season, were imbedded in paraffin melting at 54° C., sectioned and mounted in the usual way. The youngest tubercles scarcely turn brown in the fixing fluid, but older ones become brown or almost black. In any case I transferred the slides, after the paraffin had been removed by turpentine, to a solution containing one part commercial peroxide of hydrogen in twenty parts 80 per cent. alcohol. In this solution they remained a half hour, or until the sections were no longer in the least brown, and were then run down into water for staining.

The method of staining which I found most useful is a combination of Flemming's well known and now very popular triple stain—anilin safranin, anilin gentian violet, and orange G.—with Ehrlick's method of staining cover-glass

preparations of bacteria. The stains were made up according to the directions given in Humphrey's Zimmermann's Botanical Microtechnique (p. 186), and were used as Hof (1898) directs, except that after the sections had been for not more than two minutes in the gentian violet solution they were rinsed with water and placed for a half hour or longer in Gramm's iodine solution to differentiate the bacilli and the infection threads from the cytoplasm. Hof says that the sections may be left from two to three minutes in the anilin gentian violet. I often found this quite too long, and had difficulty in washing out enough of the violet without taking out the safranin also. One minute is usually long enough for these tissues. Washing off the Gramm's iodine with water, the slides were then allowed to remain for one to two minutes in staining bottles containing orange G; they were then washed with absolute alcohol so long as gentian violet came off abundantly or needed to be removed (as shown by microscopic examination), were cleared in clove oil, and mounted in xylol balsam. I decidedly prefer clove oil to xylol for clearing, as it aids in the differentiation for which this staining method is so highly prized.

So far as my experience goes, this method of fixing and staining is perfectly certain to demonstrate the infection threads and to differentiate the bacteria in the cytoplasm and in the unstained matrix of the threads. I am, therefore, somewhat at a loss to understand the difficulties reported by some authors in staining tubercles and their contents. Miss Maria Dawson (1899, p. 8) reports, for example: "For some time I made use of both hand-sections and microtome sections of paraffin material. The latter method I afterwards abandoned, however, since I found the tubercle tissues very difficult objects to stain upon the slide, and also ordinarily thin hand-sections serve better for the examination of the filaments within the cells—a point to which I wished to devote special attention." With the stains Miss Dawson used on hand-sections, and of which she speaks

favorably, I, too, failed to get satisfactory results from microtome material, butowing to the success of my own method I did not try hers very long.

II. ORIGIN AND MORPHOLOGY OF ROOT-TUBERCLES.

Although most that I can say about the entrance of the tubercle bacteria into the roots of leguminous plants has already been reported by others, I wish to describe the infection in Bur Clover (*Medicago denticulata* Willd.) from the beginning, and to discuss some of the stages in the process. I must also frankly admit that I do not know all that has been written on the subject, for the literature is copious and scattered, and I have been able to see only the papers herein referred to. I therefore bespeak lenient criticism of my acquaintance with the literature, remoteness from the centres of scientific and other work making it very difficult to secure the papers, and even references to the papers, of my subject.

How the tubercle bacteria in the soil come into contact with the root-hairs of the leguminous plants which they attack is not known. The majority of authors consider these bacteria (*Bacillus radicicola* Beyerinck, *Rhizobium leguminosarum* Frank) only slowly motile if motile at all. In artificial cultures they are usually quite motionless (Migula, 1900, p. 772). Zinsser (1897, p. 447) says they are small and actively motile. Miss Dawson (1900, p. 59) reports that in drop-cultures, a week or more old, the chains become motile, the shorter moving more rapidly than the longer, but none actively, and the motion resembles the pendulum movement of *Oscillatoria* filaments. In younger drop-cultures, containing 2.5 per cent. gelatine, I have seen this movement. The movement of the chains formed in artificial cultures may be only a feeble index of a much more active movement of the separate bacilli when these occur in natural conditions. Artificial cultures are unsatisfactory at best, and it may very well be that the tubercle bacilli are actively motile, for a time at least, in damp soil.

It is well known that many bacteria are actively motile only for a short time after division. When the conditions in the soil—whatever these conditions may be—favor the reproduction of these bacilli, the young and separate rodlets may be able to move with a fair degree of rapidity. They may possibly be stimulated into motility by the proximity of the root of some leguminous plant, and may be characteristically attracted to it by the substances diffusing from it into the soil-water (see Czapek, 1896). At first glance this seems improbable because of the very small proportion of root-hairs attacked to the total number of root-hairs formed; but on this point the following observations throw some light.

In the field of a Leitz objective III and ocular 3 I counted one hundred root-hairs on the sides of a young lateral root. Of these hairs one was infected. There must have been an equal number of root-hairs on the top and bottom of the root as it lay on the slide. The zone of hairs was about five times as long as the distance through which I counted hairs.[1] This would make the total number of hairs on this small root at least one thousand. I searched carefully, but found no other infected hair anywhere on this rootlet. The proportion of infection in this case is as one to a thousand. The rootlet examined was from a young Bur Clover seedling growing in sandy soil in the laboratory. There must have been a large number of Bur Clover bacteria in the soil used, for it was taken from a spot where Bur Clover throve last year and again this. Out of doors I fancy that the number of root-hairs would be greater in the same sandy soil than in the laboratory, for I watered my indoor material, and that outside was watered only by the rains. The number of root-hairs attacked is probably no greater, however. The proportion of one to a thousand is therefore conservative. If, then, these bacteria are motile only slowly, if at all, it is apparently the mere chance of a root-hair's growing to or very near where the bacteria are which makes infection possible. We may infer either that the

[1] This I estimated with considerable precision by means of a mechanical stage.

bacteria are not motile or move only slowly, or that it is chance which governs the infection of the root-hairs. This latter inference is encouraged by a statement of Miss Dawson's (1899, p. 21), that she saw "on a very small piece of a lateral root from one of the plants no less than twenty-seven hairs, side by side, with well grown infection tubes within them. This observation may serve to show how successful the attacks of this organism may be, provided suitable conditions can be arrived at."

To ascertain whether the number of infections in root-hairs in nature is smaller than it might be under other conditions, I tried the following experiment. Three layers of filter paper, moistened with tap-water, were laid in each of four small saucers and covered by tumblers. These were sterilized in an Arnold steam sterilizer on three successive days. A half dozen Bur Clover seeds were then placed under each tumbler on the damp filter paper. The seeds had just been removed from the little bur-like pods, dipped in corrosive sublimate solution of one to one thousand, and rinsed in distilled water which had been repeatedly boiled. The filter-papers were watered daily with boiled distilled water. In a few days the seeds germinated. When the roots had grown to an inch or so in length and had developed many root-hairs, they were watered with boiled distilled water in which had been ground healthy growing tubercles. The water therefore held tubercle bacteria in suspension. The next day nearly every hair in the field on one side of a root was found to be enlarged and twisted at the ends and showed the beginning of an infection thread. Given the contact or close proximity of the tubercle bacteria with the root-hairs, infections may take place in great numbers simultaneously, at least when the roots are very young. Whether the roots are always susceptible, or whether older root-hairs or root-hairs on older roots are susceptible, is another question. Any change in the composition of the walls of the root-hairs may affect their solubility or at least their permeability by the bacteria (cutinization?).

It would appear from another experiment that not all contacts and infections of root-hairs with tubercle bacteria lead to the formation of tubercles. Among the seedlings growing in sterilized moist chambers, I infected some with bacteria from a gelatine culture of Bur Clover tubercle bacteria. The next day showed a great increase in the number of bacteria, but the tips of the root-hairs, though bent in many instances, were not coiled in the manner usual in infections, but instead, were cut off into short sausage-shaped, often non-nucleated segments. In this way the bacteria which have entered a root-hair are excluded from the more vital parts of the root, just as gonidial cells in lichens are known to exclude the haustoria of the fungus by so dividing that only one daughter-cell contains any part of the haustorium which has penetrated the mother-cell (Hedlund, 1892; Peirce, 1899). Once given the contact with the bacteria, the root-hairs can become infected; but these infections may be resisted by the leguminous plant by cutting off the infected parts.

I am by no means ready to attach especial weight to the result of this experiment for the following reasons: First, I did not repeat the experiment, important though it would be to prove that the root-hairs do cut off the infected portions; second, this result followed the infection of the sterile root of a seedling, not by bacteria suspended in water but by stroking the root with a platinum needle which had been dipped into a culture of the bacteria. By this means not only bacteria but also their accumulated products in the culture-medium were put upon the root. It might well be that these products, rather than the bacteria themselves, so irritated the root-hairs that they segmented as above described. It would be interesting to follow this matter to a decisive conclusion, but it was not possible at the time to do so, and this point was not directly connected with the main object of this investigation. It may be that by similar means the root-hairs, and thereby the roots, of other than leguminous plants resist and escape infection by the bacteria which so characteristically affect the

leguminous plants growing in the same soil and under the same conditions. This possibility has likewise been the subject of experiment.

Returning now to the question of the motility of the nodule bacteria, we see that the experiments just described indicate that [illegible] takes place in a manner that when many bacteria are brought directly into contact with young and sterile roots, but the experiments leave the matter of the motility of the bacteria still undecided. The behavior of the bacteria in artificial culture is inconclusive, and apparently we cannot now imitate the conditions which prevail in the soil.

Figures 1–3 show root-hairs of Red Clover plants infected by nodule bacteria. Figure 1 shows the lower and larger of the two hairs in fig. 2 more highly magnified. Figure 3 is a hair on another plant drawn with the same magnification as fig. 2 (× 300). In these figures, as in those of Frank, Prunet, and many others, it is noticeable that on the concave side of the curved tip of the root-hair there is a small mass of bacteria, this mass being continuous with the line or strand of bacteria extending through the hair. The wall of the hair seems intact and uninjured except where the small mass of bacteria is. At this point there is no appearance of the wall. The wall may be actually perforated, though to see this with the mass and the strand of bacteria in place would be very difficult or impossible even in very thin sections. It is much more probable that the wall is merely softened, the cellulose digested at the point where the bacteria are—a soft place large enough in area to permit the bacteria to enter either by active locomotion or by the formation and growth of new cells in this direction.

The little group of bacteria on the surface and near the tip of a root-hair is very often at the point of greatest curvature of the hair. This curvature is due to the bacteria. The bending is the evident response to irritation. The irritation may consist in the softening and partial solution of the cell-wall by enzyme formed by the bacteria—a mechanical irritation—or in the stimulation of the cell by the same

or other substances. In the one case we have a traumatropic bending, in the latter a chemotropic. (See Spalding, 1894.) This bending is entirely different in appearance and distinct in cause from that which carries the root-hairs closely around particles of soil. This last is due to irritation by contact (thigmotropism) and by water (hydrotropism).

Since the majority of infected root-hairs show the bending at or near the tip, as shown in figs. 1–3, we may infer that the bacteria enter uninjured hairs which are able by growth curvatures to respond to mechanical or chemical stimuli. If the hairs were broken, the ability to respond, and the responses (curvatures) would be greatly lessened, and instead of a short, close spiral (figs. 1–3), or a pronounced bend, we should have little or no curvature. The curvature of a broken hair is doubtful, and for mechanical and physiological reasons certainly difficult to understand.

The roots of young Bur Clover grown in sandy soil in the laboratory showed very few broken hairs when I dug up the plants to search for infected hairs. The soil was very friable, but even then I expected to find more hairs broken as the result either of taking up the plants or of their growth in the soil.

It would appear, then, that these bacteria are able to soften or dissolve cell-wall, and when they come into contact with a root-hair, enter it, whether it is broken or not. The very slow movements, which are all that most observers report having seen in these bacteria, their ability to soften or dissolve cellulose, the small number of infected hairs, and the small number of broken hairs, make Fischer's graphic description (1897, pp. 91–2) of how the infection of leguminous plants takes place at least doubtful though no less graphic and interesting. He says: "Die feinen Wurzelhärchen einer jungen, noch knöllchenfreien Leguminosenpflanze schieben und drängen sich überall zwischen die Bodentheilchen ein, um hier Wasser und mineralische Salze aufzunehmen, ja sie scheiden sogar besondere Stoffe aus, um die Erdteilchen, mit denen sie dicht verkleben, zu lösen. So wird schon die unverletzte Oberfläche der Wurzeln

chemotaktisch wirkende Stoffe vielfach absondern. Dazu kommen noch zahlreiche verletzte Wurzelhaare oder andere leichte Wunden der Wurzel, die anlockend auf Knöllchenbakterien wirken werden, wenn diese in den wassererfüllten Räumchen zwischen den Bodentheilchen herumschwärmen. Wovon hier die Bakterien leben, bedarf noch weiterer Untersuchung, denn sie müssten hier natürlich mit bescheideneren Kohlenstoff- und Stickstoff quellen vorlieb nehmen als in der Reinkultur mit Asparagin und Zucker. Gerade solche Stoffe, besonders das chemotaktisch sehr wirksame Asparagin ist in den Keimpflanzen der Leguminosen stets reichlich enthalten und wird bei jeder Verletzung der Wurzel hervortreten. So konnte ihm wirklich die Rolle des Anlockungsstoffes fur die Knöllchenbakterien zufallen, die in ein aufgerissenes Wurzelhaar genau so einschwärmen würden, wie in eine mit Asparagin gerfüllte Kapillare." Fischer's conviction, expressed at length and supported as well as possible by example and argument (l. c., pp. 131–2), that bacteria do not enter uninjured plant-cells and hence cannot produce disease by being parasitic on or in plants, is probably responsible for this statement, which seems to me the opposite of correct. If it can be shown that bacteria of any one species penetrate the cell-wall of healthy uninjured plants, producing unusual growths therein, Fischer's contention that there are no bacterial diseases of plants breaks down. On this question Smith (1901) takes issue with Fischer, and seems to prove that there are bacterial diseases of plants.

To help himself over the unavoidable difficulty of the tubercle bacteria entering the roots of plants, Fischer says (l. c., p. 92): "Ja es scheint sogar, als ob die Leguminosen durch Auflockerung der Zellwände an manchen Wurzelhaaren u. s. w. die Anlockung der bacterien vorbereiten. In dicht gedrängten Zügen dringen sie von der Oberfläche der Wurzel in deren Inneres vor, wobei ihnen wiederum die Leguminose den Weg zu ebnen scheint dadurch, dass sie die schwer durchdringbaren Zellwände etwas auflockert." Such a loosening or softening (Auflockerung)

of the cell-walls is well enough known in other cases; but it is not the host which softens its own walls in order to facilitate the entrance of a foreign organism, but rather the foreign organism which, by enzyms secreted by itself, softens or dissolves the walls of its host which lie across its path of growth.[1]

Having entered the root-hair by softening or dissolving a small portion of the cell-wall, and moving or growing through this, the tubercle bacteria multiply rapidly, forming a thread-like zooglœa from the infection spot along the hair into the epidermal cell of which the hair is a branch (figs. 2 and 3). From the epidermis the infecting zooglœa grows fairly straight into the underlying cortical parenchyma. Figure 4, drawn from one of a series of thin microtom sections stained as previously described, indicates the course of the infecting strand (purple). This course is nearly, though not quite, straight toward the central cylinder of the root, for within a series of five or six sections—a distance of 20–30 μ—the infection thread was traced from the base of the root-hair (*r. h.* in fig. 4) to one cell (10) in the layer next to the endodermis of the central cylinder. The cells in this layer are distinguished from the cells of the cortical parenchyma by somewhat larger and denser nuclei. This layer is the one from which the lateral roots arise. The direction of the infection thread—which is solid, and is incorrectly termed infection "tube"—is too regular not to encourage one to suppose that the course of the growing strand of bacteria is determined by attraction exerted by the host-cells upon the bacteria. This then is chemotropic growth of the strand or, if the bacteria are motile in the cells, chemotactic movement of the bacteria. The course of the thread is toward the conducting tissues of the host. This is similar to the growth of the haustoria of Dodder, *Cassytha*, *Viscum*, *Phoradendron*, and other phanerogamic parasites (Peirce, 1894; Cannon, 1901). The growth does

[1] See De Bary ("Morphology and Biology of the Fungi, Mycetozoa, and Bacteria," Oxford, 1887) and many others as to this in fungi, and Peirce (Annals of Botany, 1894) as to Dodder.

not extend into the central cylinder and the conducting tissues, so far as I have seen. Instead, in the layer of cells just outside the endodermis of the root, division takes place in the cell into which the infection thread has penetrated and in the cells adjacent to it. The daughter-cells grow, repeated divisions and growth follow, and there arises a conical mass of cells which are somewhat larger, and which contain more protoplasm, than the adjacent cortical parenchyma cells. This conical mass is the young tubercle. At first all of its cells are merismatic, but later the divisions become more and more limited to the cells near the rounded apex of the blunt cone. Thus a regular cambium is differentiated in the tubercle. This cambium, as shown in fig. 7, lies near the tip of the tubercle, and forms a bowl-shaped or shallow thimble-shaped layer.

The growing tubercle pushes out the overlying cortical parenchyma and epidermis, forming an increasing swelling on the side of the root. Cortical parenchyma and epidermis, at least for a time, nearly keep pace with the growth of the tubercle. Thus, although the cortical cells are compressed somewhat, the epidermis is not ruptured, and the tubercle does not burst out of the side of the root as a lateral root does.

The layer of cells which, on infection, gives rise to the tubercle, forms new cells not only centrifugally but also centripetally, so that by these new and growing cells the tubercle is pushed outward, away from the central cylinder. In this way the cylindrical mass of the root itself is kept fairly uniform. An older tubercle appears to be attached to the root. Only by tracing its development can one see that it originates internally. The course of development was only very imperfectly traced by Schneider (1893); hence his bold and erroneous statement that "tubercles seem always to develop exogenously."

Frank (1890, p. 70) states that the tubercles are new organs formed and well nourished by the plant. He compares them with galls which are formed by plants at the points attacked by parasites (insects, worms, etc.), and

adds, to strengthen his comparison, "die Wurzelknöllchen sind kein Organwelches der Leguminose ursprünglich eigen wäre, ebensowenig wie dies bei den anderen Pflanzen der Fall ist, sondern eine erst von dem Rhizobium angeregte, dann aber selbst aufgebaute Bildung." Further on, he says that he has repeatedly seen lupines, cultivated in sterilized and uninfected soil, which formed swellings on the roots closely resembling young tubercles, but showing neither infection threads nor any traces of the cell-contents characteristic of true root-tubercles. He accounts for this not on the ground of slight infections producing only abortive tubercles, but on the hypothesis that the lupines, accustomed for thousands of years to symbiotic existence with the tubercle bacteria, have so firmly acquired the habit of forming tubercles that they begin to form them even before and without infection. On these points I wish again to call attention to the fact that the tissues of the tubercle originate from the same layer of cells as gives rise, by similar divisions, to the lateral roots (see figs. 4, 5, 6). When one compares a very young mass of tubercular tissue, still enclosed in the cortex of the root, with a very young lateral root also still enclosed in the cortex of the root, the resemblance between the two structures is strong. Figures 5 and 6 show this. Figure 5 is a diagram of a section in which a tubercle and a lateral root are growing side by side and from the same layer. In the figure the tubercle is to the left, the lateral root to the right. Figure 6 is a drawing of tubercle and lateral root on a larger scale, the root to the right, the tubercle to the left. In the tubercle some infection threads show. The tubercle has the same form as the root, but shows no differentiation among its cells. The lateral root already shows a differentiation of dermatogen and there is a foreshadowing of the vegetative point. Central cylinder and periblem are not yet distinguishable. The cells of the tubercle are larger than those of the lateral root, but the nuclei of the tubercle cells are not proportionally larger and most of them are actually no larger than those of the lateral root.

As tubercle and lateral root grow, the resemblances between the two decrease and finally disappear altogether. The tubercle has no cap[1] and no central strand of conducting tissue. The tubercle cells differentiate into definite tissues more slowly than do those of the lateral root, but near the tip of the older tubercle there is a mass of meristematic cells similar to the growing-point of a lateral root (see fig. 7). This meristem forms cells forward and backward as does the growing-point. The central mass of the tubercle is proportionally much larger than the central cylinder of the lateral root, but it is wholly undifferentiated. The cortex of the tubercle contains vascular bundles, small and separated from each other by considerable spaces of parenchyma (see fig. 7), and is enclosed by layers of cork-cells. These may and usually do become powdery on the surface and rub off as the tubercle forms, just as the cap cells do from the tip of a root.

In point of origin and in their earliest growth, tubercle and lateral root are similar. In subsequent growth they are more and more dissimilar. Morphologically, then, the root-tubercles are lateral roots. Though called into activity by very different causes, the cells of the pericycle give rise by division to masses of cells which, on the one hand, develop into tubercles, on the other develop into lateral roots. In the one case we know the stimulus which causes the tubercle to form. It is the infection of the root-cells down to the innermost layer of the periblem by bacteria. Do lateral roots form as the result of external stimuli or are they the effects of causes internal to the plant? The latter is the less likely from the fact that the size, number, and position of lateral roots varies in plants of the same species according to the soil, to the number and kind of other plants living in the same soil, to the distribution of moisture and other matters in the soil, and to a great number of other factors not now recognized. This subject merits investigation. As

[1] According to Life (Botanical Gazette, April, 1901) the roots infected by *Anabæna* and certain other organisms in certain species of *Cycas* also have no caps, yet he unhesitatingly describes them as roots.

plants are studied with a view to ascertaining the effects of each factor in the environment, it will become more and more evident that many of the effects which are now attributed to internal causes, lumped together under the name inheritance or distributed among the various functions of the organs of the body, are the reactions of the parts to stimuli exerted upon them from outside. If the formation of lateral roots by the division of the cells of the pericambium should prove to be the result of external stimuli, it will be found that these stimuli operate upon the cells immediately concerned.

The diverse development of tubercle and lateral root, the result of the persistence of the different stimuli which called them into existence, obscures the common morphology of the two organs so that it is only natural that Frank should have called the tubercles new organs. If the tubercles were the result of hypertrophy indiscriminately among the cells of the cortex of the root, as may be the case in several of the species which Frank studied and described, this would be no evidence that the tubercles are morphologically roots. But in Bur Clover the case is clear. Doubtless the same can be made out in other leguminous plants when the origin of the tubercles is studied in properly stained microtome sections of young roots. I have not examined Frank's *Lupinus*, bean, pea, etc., for it is a matter costing much time and patience to find just the right stages, and I preferred to study my own plants and to leave a review of the origin and structure of the tubercles of the plants studied by Frank to a later time or to others interested.

In Bur Clover, at least, I think I have advanced strong evidence against Frank's contention that the tubercles are new organs. As to his hypothesis, involving inheritance as one of the factors in their origin and development, that lupines grown in sterilized and uninoculated soil may form small enlargements like young tubercles, I think my demonstration of the common origin of tubercles and lateral roots also has some significance. Assuming that the soil remained sterile, which is not wholly probable, the plant might begin

to form lateral roots which, for some unknown reason, aborted while still in the cortex of the mother root. Lateral roots are known to do this, and if the abortion took place early enough, the root character of the new formation might be lost if it had already developed. It seems to me much more probable, however, that both causes were in operation; that the soil did not remain sterile; that the plants were infected by so few or so feeble tubercle-bacteria that the tubercles stimulated to begin to form aborted because the infection was not strong enough. If the leguminous plant, or its separate cells, and the bacteria are parasitically associated, the plant would resist the entrance and growth of the bacteria, and would be much more likely to succeed in this if the attacking bacteria were few or feeble. Overcoming the bacteria, the stimulus to tubercle formation ceases, the tubercle remains rudimentary. That infection of sterilized soil by the tubercle-bacteria is possible, and even difficult to avoid, is known to all who have worked on the subject. This, then, rather than inheritance, accounts for the rudimentary tubercles which Frank describes.

The bacteria in the infection thread, which grows through the root-hair and the cortical parenchyma cells of the root to the pericambium layer, multiply, but they multiply most rapidly in the infected cells farthest from the surface of the root. New threads form, which grow out into and infect the cells of the mass of new cells composing the embryo-tubercle. Thus a majority of the cells in the young tubercle contain bacteria.

Though infected cells do divide (see pp. 322–323), they probably divide less often than the uninfected cells. The primary infection is in a nearly straight line from the root-hair inwards. The infection of the daughter-cells composing the embryo-tubercle is accomplished by branching infection threads growing in fairly straight lines radiating from the base of the tubercle. In this way the cells near the base of the growing tubercle are most infected, those near the tip least. It may be in consequence of this that the cells at and near the tip of the tubercle retain their

merismatic quality, and that they form the bowl-shaped layer or layers of meristem which continue the growth of the tubercle (see fig. 7). The infection threads grow out toward the tip of the tubercle (fig. 10), but the meristem continues to form new cells between itself and the cells containing bacteria and infection threads. By the layer or layers of uninfected daughter-cells the meristem protects its own cells from infection. Perhaps because they escape infection, they retain their ability to divide. If they can be prevented from forming a sufficient number of daughter-cells to enable them to escape infection, what will be the result?

To answer this, and some other questions, I tried the following experiment. I imbedded young tubercles on growing roots of potted plants of Bur Clover in plaster of Paris, according to the method devised by Pfeffer (1892) and used by his pupils (Newcombe, 1894, Richter, 1894, etc.). The roots were disturbed as little as possible and were put back carefully in the soil as soon as the plaster had been applied to the tubercles. The plants were growing in coarse sandy gravel, so that it was not difficult or dangerous to lift out one or more roots when the soil was well loosened by being soaked with water. After the lapse of twenty-nine days, I again took up the roots, cutting them off for careful examination. Two tubercles had been firmly held at the base by the plaster, but had broken the casts sufficiently at the tips to grow fresh and pink out of their investments. New tubercles seemed to have been formed since the roots were operated upon. One tubercle, which had stayed in its cast, was taken out and sectioned by hand. It was evidently dying. The bacteria in it seemed active, but were fewer and much smaller than in unconfined tubercles. *There was no meristem;* the cells of the tubercle were in their definitive condition; there were smaller quantities of starch in this than in ordinary tubercles. What was true of this tubercle was equally true of others which had failed to break and grow out of their casts.

It would seem, then, from the results just described, that the tubercle meristem is preserved from loss of its merismatic qualities by escaping infection, and that the presence of bacteria in the cells ultimately costs them their power of division (see pp. 322–323). The meristem near the tip of the tubercle is a survival, as is the vegetative point at the root-tip, of the merismatic cells which constitute the tubercle and the root in their embryonic condition.

The result of imbedding an infected tissue, a root-tubercle, in plaster, is different from that described by Newcombe (1894), who worked on healthy plants. He says that the external mechanical resistance causes developing cells to attain their definitive condition more slowly, continues the merismatic power and activity of the cambium cells, and prolongs the life of such cells as ordinarily die early. Newcombe worked on healthy plants, and his results exhibit the effect of pressure only on growing cells and organs. In my experiment the pressure which checked the growth of the tubercle-cells may not have mechanically affected the bacteria. Since the bacteria and the cells of the tubercle are competitors, the plaster investments handicapped the latter to such an extent that the ultimate results of bacterial activity appeared earlier than in unconfined tubercles, the bacteria gaining an advantage. To the action of the bacteria rather than of the plaster cast are due the early loss of merismatic power and the early assumption of their definitive condition by the cells of the tubercle.

III. The Form and Distribution of Root-tubercles.

The form and distribution of root-tubercles merit some discussion, and since those of Bur Clover are typical I will continue to describe them. The tubercles grow both in length and in thickness at the ends, not at their bases, and thus become club-shaped. They may and often do branch. The growth takes place solely at the tip, because the only meristem is there. The tubercle tissue is supplied with food through vascular bundles which are neither large

nor numerous but adequate for a time. A lateral root grows both at the tip and throughout its length, thickening and elongating. In this way its cylindrical form is maintained. The lateral root contributes food-materials and water in increasing amounts to the plant which forms it. The tubercle receives food from the plant. Perhaps it contributes also to the nutrition of the plant. Experiment so far seems to justify this belief. But if the tubercle were altogether beneficial and increasingly so, one would suppose that it would grow at the base, by secondary thickening, as well as at the tip, by primary growth, in order through increasing conducting tissues to contribute more and more to the nutrition of the plant. The absence of such secondary growth and the ultimate fate of the tubercle—dying and being cut off—suggest that the leguminous plant limits as far as possible the supply of food to the tubercle, and finally stops it. Herein we have another item of evidence against Frank's hypothesis that the leguminous plant encourages tubercle formation. It does not cut off the tubercle immediately; the irritation which results in tubercle formation is too great and the osmotic demand for food is too strong to be resisted at once by the plant. Only after a time is tubercle growth checked—perhaps by the remoteness of the tubercle meristem from the source of food-supply—and later still, the tubercle is cut off.

The tubercles may be rosy pink at and near the tip, creamy white elsewhere, nearly or quite the same shade as the roots bearing them. Later, the oldest tubercles, those nearest the surface of the soil, may branch, taking on a flat, though thick, fan shape. By no means do all of the tubercles branch. Those very near or almost on the surface of the soil do not, and in the lower half of the infected portion of the root-system I have seen almost no branched tubercles. The difference in age between the branched and unbranched tubercles in the same or the adjacent layers of soil is not sufficient to account for the difference in form. The branched tubercles are the first ones to lose the plump and healthy appearance of active life; they grow thin and

shriveled. They have grown fast, attained maturity early, and they die young. The reasons for this are probably two: First, in the upper layers of soil, which are certainly best aërated, the bacteria in the tubercles obtain the uncombined nitrogen which they absorb and fix (Mazé, 1897) more readily and more abundantly than those in tubercles farther down; hence, second, they grow and multiply more rapidly, the tubercle-cells are irritated proportionally. Because the rapid growth and multiplication of the bacteria, and, probably as a consequence, of the tubercle-meristem cells also, are not uniform, branching occurs as a result of some parts of the tubercle growing faster than others. The greater activity of the bacteria and of the host-cells in these branched tubercles is not accompanied by adequate, much less proportional, growth of the base, and of the conducting tissues in the base, of the tubercle. These tubercles sooner cease to receive as much food from the leguminous plant as they need, and hence are the first to die. Since these branched tubercles are the largest and contain most bacteria, one would suppose they would benefit the plant more than the smaller ones (if any tubercles are beneficial), and that they would be best supplied with afferent and efferent conducting tissues, as indicated by the proportional size of their bases. This is not the case.

The vertical distribution of tubercles on the roots has been reported by Frank (1890, pp. 22–3). The greatest number and the largest tubercles occur on the lupine within seven centimeters of the surface of the soil, and there is a rapid decrease in both number and size as the depth increases, till below fifty-three centimeters none was found. The distribution on Bur Clover roots corresponds. The strictly aërobic character of the tubercle bacteria, as shown by artificial cultures, accounts for this, but the relations of these organisms to the uncombined nitrogen of the air as well as to the oxygen should be borne in mind when these bacteria are said to be aërobic. The distribution of air in a soil varies with the nature of the soil—a well drained gravelly soil being well aërated to a greater depth than a

heavy, compact, clay soil. It also varies with the tillage. I have found tubercles much lower on the roots of Bur Clover growing on a heap of gravel than in an undisturbed and compact clay. Mechanical reasons are insufficient to account for this. The only inference to be drawn is that the bacteria are limited in their natural distribution to those soils and those layers of soil which contain considerable volumes of air, for only there will they find enough oxygen and nitrogen for their needs.

It is the general habit of leguminous plants to send their roots fairly deep into the soil. In a natural field, or one returned to a state of nature, where the soil is covered by a mixed vegetation, it is found that different plants send their roots to different depths. In this way the resources of the soil are more perfectly exploited by the plants and destructive competition is avoided. But it is to be noted that of those plants which send their roots deeper, many are members of the Leguminosæ. Is this merely to escape competition with other seed-bearing plants, or to reach a more abundant and constant supply of water, or to escape the attacks of the bacteria which cause them to form tubercles? It is mainly the Leguminosæ which are successfully attacked by tubercle bacteria, and they, as a rule, send their roots fairly deep into the soil. Furthermore, the number of roots increases with the distance from the surface. It would appear not inconsistent with the evidence so far obtained, to suppose that the habit of the Leguminosæ of sending their roots deep, and of causing them to branch copiously only after they have reached some depth, is one means which these plants have of avoiding the attack of tubercle bacteria.

Alfalfa or Lucern (*Medicago sativa* Linn.) has notoriously long roots. They are reported to go down to the permanently water-bearing levels of the soil. The best chance for examining these roots would be offered when a well is being dug where Alfalfa is growing. I have not been so fortunate as to have such a chance, but digging around Alfalfa plants growing as weeds in a grass field

shows that below a depth of twenty centimeters from the surface the number of tubercles decreases rapidly. The roots of this plant are perennial, and the new roots each season are most of them formed so far below the levels in which tubercles ordinarily occur on the roots of leguminous plants that this plant should form a good test object of the vertical distribution of tubercle bacteria in the soil. Compared with such annuals as Bur Clover, a member of the same genus, there were far fewer tubercles on Alfalfa roots than on Bur Clover at the time that I dug around Alfalfa (December). There were young roots near the surface as well as further down, but the greater number of young roots must have been formed far below where I reached by digging, for there were not enough young roots above to meet the needs of the plant. Alfalfa and many other perennial Leguminosæ, may therefore form the majority of their new roots each year so deep in the soil that they cannot become infected. As the tubercles are not perennial, whatever advantage may accrue to the perennial leguminous plant by its association with bacteria would be limited in time and quantity to the early life of the individual, when its roots were all in the layers of soil containing active tubercle bacteria. The question is well worth study.

IV. The Structure of Root-tubercles.

The structure of a tubercle is shown somewhat diagrammatically in fig. 7. This is a sketch, at a magnification of thirty-five diameters, of a section of a young and still growing tubercle. The section is parallel with the long axis of the tubercle and at right angles with the root. The cambium of the tubercle lies between *a* and *b*, parallel with and in the broken curved line. This meristem is composed of two or three layers of cells. Those toward the periphery of the tubercle as well as those toward the center divide, the cells toward the periphery differentiating rapidly into cells which round off from one another and form the powdery, cap-like tissue which wears away but protects the meristem within just as the root-cap protects the

growing-point of the root. The cells toward the center differentiate somewhat diversely according as they become infected by bacteria or continue free from them. The uninfected cells remain comparatively small, and present the characters of ordinary parenchyma cells, the protoplasm becoming vacuoled and containing numerous starch grains. There may be several vacuoles in these cells or one traversed by strands of cytoplasm.

The infected cells grow larger and in their definitive condition are from half as large again to twice as large as the uninfected cells. This increase in size may be attributed to one of three causes: First, to the stimulation of the protoplasm by the bacteria and the substances produced by them in the cells; second, to the actual irritation (inflammation) of the protoplasm; and third, to the increased pressure set up in the cell by the rapidly multiplying bacteria. By the plaster of Paris method we can test the relative value of two of these influences; the third must be determined by ascertaining microscopically the actual condition of the protoplasm of infected cells. On imbedding the young tubercles of growing roots, as above described, the pressure normally or abnormally developed in the cells will be resisted by the plaster, the cells expanding against the plaster will be subjected to compression. Nine days after the tubercles were enclosed in plaster, I opened the casts and sectioned the tubercles by hand. There were many more starch-grains in the uninfected cells than in ordinary tubercles; I saw no infection threads (I did not fix and use the triple stain previously described and hence infection threads might have been present which escaped my notice), the bacteria and bacteroids were smaller, and the general appearance suggested that the leguminous cells were better able to bear confinement than the bacteria were. Increase in size both of the tubercle and of its component cells being prevented by the plaster investment, the bacteria have no increasing space in which to grow, and continuing to multiply, for a time at least, they remain

small, in consequence consuming less of the food provided for them by the leguminous plant. Starch can therefore accumulate in the uninfected cells from which the infected cells are osmotically supplied with food. Depositing the non-nitrogenous food in solid form (starch) of course reduces the turgor pressure in the cells of the tubercle and thereby reduces the resistance to the plaster investment. The component cells of the tubercle enclosed in plaster of Paris remain smaller than in unconfined tubercles, the infected and uninfected cells being more nearly equal in size.

Enclosing a tubercle in plaster may diminish the stimulus or inflammation produced in the cells by the bacteria, but as the bacteria survive and multiply there can be no great diminution of their chemical effect on the cells in which they occur. The physical result, pressure, is much more affected by the plaster investment. Since the infected cells remain more nearly the same in size as the uninfected ones, the inference is plain that the excessive increase in size of the infected cells is due to increased pressure in them.

The infected cells, as shown by fig. 8, are thin-walled and contain only one large vacuole. This is not traversed by cytoplasmic strands. The quantity of bacteria may vary in infected cells, and with this there is a corresponding variation in the appearance of the cells. Thus, fig. 8 shows a typical infected cell in which the bacteria have multiplied enormously, while fig. 9, magnified about one-third larger, represents a cell in which there are comparatively few bacteria, most of which are at the point indicated by the line from *B*. In the cells containing relatively few bacteria there may be some starch-grains, as indicated by the line from *S*. In unstained hand-sections, the degree to which the older parenchymatous cells in the central part of a tubercle are infected is indicated at a glance by the amount of starch in the cells, the cells with the average amount of bacteria containing no starch-grains, the cells with no bacteria containing many starch-grains, the cells with few bacteria containing starch grains in inverse proportion to

the number of bacteria. The cells with few or no bacteria receive more non-nitrogenous food than they consume. The excess they deposit in solid form as starch. The cells with many bacteria presumably receive at least as much non-nitrogenous food; but either they themselves or the bacteria in them consume this so that there is no excess to deposit.

From the cells toward the center of the tubercle the new cells formed by the tubercle meristem are infected by means of infection threads running fairly straight toward and into the daughter-cells of the meristem (see fig. 10). This figure, magnification two hundred, was drawn from a thin microtome section of a young tubercle, and is colored as nearly as possible like the cells of the preparation. The preparation was stained, as previously described, by Flemming's triple stain, Gramm's iodine solution being used after the anilin gentian violet in order to differentiate the strands of bacteria. The cell walls are drawn in black, though they were only very faintly stained and of course were not black. The cytoplasm is brownish yellow from orange G., the nuclei a somewhat deeper shade of the same color, the nucleoli red from anilin safranin, the infection threads purple from anilin gentian violet. In a fresh preparation, whatever starch grains are present in the section are stained the usual color by the iodine, but this color is fugitive. The arrow to the left in this figure (fig. 10) shows the direction in which the tubercle meristem lies. It is evident from this figure that the infection threads run very definitely toward the new cells formed by the merismatic layer. Since all the food of the tubercle cells comes from the opposite direction, from the root, there must be some other directive influence than this food to cause the infection threads to grow so definitely toward the tip of the tubercle. This influence must come from the cambium or from its daughter-cells, and must consist in the substance or the products of these cells rather than in the food supplied to them. If we are to assume any chemotactic influence, it must be exerted by some diffusing substance or substances and

hence must be by the products and not by the living substance of the meristem and its daughter-cells. The direction of growth of these infection threads cannot be determined by the oxygen or nitrogen (or both) of the air, for if this were the case, we should find strands of bacteria running from the central cells in all directions toward the periphery of the tubercle. This is not the case. The strands run toward the growing-point of the tubercle. In consequence, the daughter-cells successively formed by the repeated division of the cells of the meristem become infected.

Not only do the infection threads run definitely toward the growing-point of the tubercle; they also grow toward the nucleus of each cell which they enter. This statement has been repeatedly made and denied in papers on the subject of root-tubercles. In hand sections, especially if the material were not carefully fixed and differentially stained, it would be easy to find evidence in support of the affirmation and of the denial. Microtome sections, differentially stained as before described, of carefully fixed growing tubercles of the species of leguminous plants which I have especially studied, show that in most cases the infection threads run definitely toward the nuclei of the tubercle cells. This is evident in fig. 10. Figures 11 and 12 also show this. In fig. 13 are shown two tubercle cells in which the main infection thread is not directed toward the nuclei, but the lower of these two cells shows that the infection thread bends toward the nucleus. In the next section of the series (not figured) a branch runs from the main infection thread to the nucleus. In the cell shown in fig. 11 the infection thread is divided, one part running beneath, the other above the nucleus. For the sake of clearness this upper part of the thread was omitted in drawing. The nucleus of the upper cell in fig. 13 was not in the plane of the section. An adjacent section (not figured) contains this and has a branch of the main infection thread running to it. There must be some reason for this definite growth of the strands of bacteria toward the nuclei of the cells which

they enter.[1] The effect on the nuclei is marked, as will be shown presently.

The changes which take place in infected cells as they develop are indicated in figs. 14 *a*, *b*, *c*, *d*. These represent successive cells from the meristem backward toward the center of the tubercle, which is shown in fig. 7. A part of the cambium layer at *a–b* in fig. 7 is shown at *x–y* in fig. 14 *a*. This series is stained by Flemming's triple stain, but Gramm's iodine was not used. For this reason the bacteria and infection threads are not differentiated. The magnification of figs. 14 *b*, *c*, *d* is 300, that of fig. 14 *a* is 270, hence 14 *b* and 14 *a* do not meet exactly. The figures in the series had to be drawn separately by reason of the limited field of the objective which was used to give the necessary magnification.

Two cells of the tubercle cambium are shown at *x–y*. Recently formed daughter-cells lie toward the outside as well as toward the center of the tubercle. The cell *z* is already beginning to show the effects of infection, vacuoles of considerable size, which later become confluent, forming in the cytoplasm, and a distinct vacuole, like a halo, appearing around the nucleolus. As shown by the cells further toward the center of the tubercle, the nucleolus is the first part of the nucleus to be evidently affected by the presence of tubercle bacteria in the cell. It is the first part of the cell to decrease in size and to disappear. If the nucleolus is in fact an accumulation of food in the nucleus, one would expect it to disappear, to be consumed, whenever there arose a special need of food in the nucleus, or even in the cell as a whole perhaps. Furthermore, the

[1] W. Magnus (1900) discusses the endotrophic mycorrhiza fungus of *Neottia nidus avis* L. in this connection, stating (pp. 7-9) that the hyphæ of this fungus do not grow toward the nucleus of the cell with any regularity, but that in many other parasitic fungi the hyphæ do grow toward and around the nucleus, in some cases, however, with no greater regularity than toward and around starch-grains or other solid contents of the cell. He adds (p. 61): "Dass sich parasitäre Pilze mit ihren Haustorien oft an den Zellkern legen und sich in seiner Nähe eigenthümlich verzweigen, gestattet keinen Rückschluss auf die Bedeutung des Kernes als Nahrungscentrum der Zelle." To conclude, because fungus hyphæ or strands of bacteria grow toward the nucleus of a cell which they have entered, that the nucleus is the center of nutrition of the cell is illogical; but it may well be in these cases that the nucleus contains or produces substances which nourish and attract the parasites.

greater solubility of the substance of the nucleolus than of the substance of the nucleus and cytoplasm would make it the first to disappear under the attack of a parasite.

In fig. 14 *b* two characters of the cells are especially noticeable: the nuclei are becoming disorganized, cavities taking the place of the nuclear substance; and the cytoplasm is no longer so dense as in the younger cells shown in fig. 14 *a*, nor does it stain so deeply with the orange G. The degeneration of the nucleus is especially marked in the cells *c*, *i*, *h*. These cells have lost their nuclei, except the last traces of acromatic substance, before they have themselves grown to their full size.

The two upper cells of fig. 14 *c* have reached the maximum size of infected cells. It is impossible to say whether they have grown to this size after their nuclei were reduced to the condition depicted or while their nuclei were being destroyed. I cannot say whether the smaller cells *c*, *i*, *h* of fig. 14 *b* would ever have grown to the size of the upper two in fig. 14 *b*, but the unusual growth of infected cells is due to the bacteria, and it is the bacteria which also destroy the nucleus or cause it to be destroyed. In the upper cells of fig. 14 *c* the cytoplasm is no longer the clear color produced by orange G. This brownish yellow color is dulled by the purple of the now extremely numerous bacteria. The smudgy appearance of the cytoplasm of the cells in 14 *d* is due to the mixture of the purple stain of the bacteria and the brownish yellow of the cytoplasm, an effect which is very striking in the preparations. The cells of fig. 14 *d* also show the greatly reduced nuclei and the large central vacuoles. Fig. 8 represents a cell from near the center of the tubercle shown in fig. 7. The cytoplasm is crowded with bacteria, the central vacuole is very large, the nucleus is reduced to an elongated lumpy mass as seen in section, or to a thin lumpy plate in the entire cell.

When infected cells contain any considerable number of bacteria, they cease to be able to divide. Freshly infected daughter-cells of the cambium layer do divide, as fig. 15 shows. One is inclined to say that the disturbing, if not

already destructive, effect of the bacteria upon the nucleus and other parts of the cells causes them to lose their power of division. As has been previously pointed out (p. 311), between the tubercle meristem and the infected cells there normally lie two or more layers of uninfected daughter-cells. These divide also and grow. By this means the meristem cells are kept from infection. When, however, the growth of these daughter-cells is prevented, and the rate of division of the meristem cells is reduced by such mechanical pressure as a plaster of Paris investment of the whole tubercle, the infection progresses to and into the merismatic cells and they presently lose their merismatic character and take on their definitive form.

One result of the division of infected cells is the production of new cells already infected and therefore not requiring the entrance of infection threads. As shown by Hedlund (1892) and myself (1899), the gonidia of lichens frequently divide in such directions as to exclude the haustoria from some of the daughter-cells. No such result can follow when a cell contains a number of minute parasites distributed in its cytoplasm. The cell may divide under these conditions, but the daughter-cells share the parasites as well as the substance of the mother-cell.

The figures above referred to, especially figs. 14 *a*, *b*, *c*, *d*, and fig. 8, plainly show that the presence of tubercle bacteria is not beneficial to the cells which contain the bacteria. So far as the relations of the bacteria and their host-cells are concerned, no one would hesitate to call the association a clear case of parasitism of bacteria in the cells. Whether it is a benefit to the leguminous plant to form tubercles, to harbor bacteria in these, to have the cells of the tubercles destroyed, and all the food supplied to the tubercle consumed, is another question. The most careful experimentation and the critical examination of the results of experiment have so far led to the generally accepted belief that the association of bacteria with the roots of leguminous plants is beneficial to the leguminous plants. I find it hard to understand how association with bacteria

which destroy the cells in which they occur can benefit the larger member of the association. The bacteria are known to fix free nitrogen from the air. By this means they form in the cells of their host-plants nitrogenous compounds which the host may use. Apparently the bacteria form more than enough of these valuable nitrogen compounds in the cells of their hosts to compensate for the extra material used in forming and maintaining tubercles the cells of which are ultimately destroyed by the bacteria. This implies a marvelous balance of profit and loss, the more remarkable since the profit apparently exceeds the loss.

One point more needs to be made clear. Miss Dawson (1899, p. 14) says that it is difficult to conceive how such strictly aërobic bacteria as these can flourish in the cells of such compact tissue as composes the tubercle. This difficulty is of her own conceiving, for do not the cells of the tubercles respire and are they not necessarily supplied with oxygen for respiration? Again, intercellular spaces in the infected tissue do occur, as figures 14 *b*, 14 *c*, 13, 12, 15, 8, and 9 plainly show. Even if intercellular spaces did not occur, as asserted by Schneider (1893, pp. 786 and 787), the existence of the living cells of the tubercle tissue would prove the presence of sufficient quantities of oxygen, if not of nitrogen, in the tissue and therefore in the cells. Unless we are to imagine anaërobic respiration for these cells, it is unnecessary to assume it for the bacteria which infest them. Fischer (1897, note 63 to p. 92), shows this clearly, and my sections reveal the presence of intercellular spaces through which a diffusion of gases could take place even if the diosmosis of solutions of the gases concerned were inadequate to supply the demand.

SUMMARY.

1. Though the bacteria which form root-tubercles on leguminous plants are usually only slowly motile, if motile at all, in artificial cultures, this proves nothing as to their motility in the soil.

2. The proportion of root-hairs infected to the total number formed is small, in one case computed to be 1:1000.
3. Given the contact or close proximity of tubercle bacteria with the root-hairs, infections may take place in great numbers simultaneously, at least when the roots are very young.
4. Infection of the root may be resisted by cutting off the infected ends of the root-hairs.
5. The tubercle bacteria enter and infect a root-hair by softening or dissolving a small portion of the wall and moving or growing through this. There is no evidence that they usually enter through broken root-hairs, and the curvatures of infected root-hairs are evidence against these hairs having been broken at any time.
6. The infection thread grows fairly straight, being chemotropically attracted, through the cortical parenchyma, from the root-hair to the layer of cells next outside of the central cylinder of the root.
7. The tubercles originate only endogenously and from the same layer as gives rise to lateral roots. We may therefore conclude that the tubercles are morphologically lateral roots, though greatly modified by the influence which caused them to be formed.
8. Tubercles form only as the result of stimulation by bacteria. Do lateral roots form as the result of internal causes or of external stimuli?
9. The growth of the tubercle is apical, the daughter-cells of a bowl-shaped terminal meristem constituting the growing part of the tubercle. There is little or no secondary growth in thickness. Because of this, the conducting tissues do not keep pace with the growth of the tubercle. The growth of the tubercle is correspondingly limited.
10. Tubercles are largest and most numerous near the surface of the soil. It is possible that perennial Leguminosæ form few if any tubercles after their roots have grown deep into the soil.

11. The presence of bacteria in the cells of a tubercle prevents the infected cells from forming starch granules. Uninfected cells do not attain the size usually reached by infected cells. The larger size of infected cells is due to increased pressure, probably also the greater irritation, in these cells.
12. The bacteria cause the degeneration and almost complete destruction of the nuclei of the cells in which they occur.
13. The infection strands grow definitely, chemotropically, toward the daughter-cells formed by the tubercle meristem, and seem also to grow definitely toward the nuclei of the cells into which they penetrate.
14. Infected cells soon lose their power of division, though not of growth.
15. The presence of bacteria in the cells of the tubercle is injurious to these cells, and the relation of the bacteria to their host-cells is parasitism.
16. It is difficult to understand how the leguminous plant as a whole can profit by an association which is injurious and finally destructive to the cells in which the bacteria occur.
17. Intercellular spaces occur in the tissues of root-tubercles. Even if they did not, it would not be necessary to assume that the bacteria live anaërobically therein, since the tubercle cells do not live anaërobically.

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BIBLIOGRAPHY.

1890. FRANK, A. B. Ueber die Pilzsymbiose der Leguminosen. Sonderabdruck aus den Landwirthschaftlichen Jahrbücher. Berlin.

1893. ZIMMERMANN, A. Botanical Microtechnique. New York. Transl. by Humphrey.

1898. HOF, A. C. Histologische Studien an Vegetationspunkten. *Bot. Centralblatt*, Bd. LXXVI, pp. 65–69.

1899. DAWSON, MARIA. Nitragin and the Nodules of Leguminous Plants. *Phil. Trans. Roy. Soc.*

1900. MIGULA, W. System der Bakterien. Bd. I, 1897; II, 1900. Jena.

1897. ZINSSER, O. Ueber das Verhalten von Bakterien, insbesondere von Knöllchenbakterien, in lebenden pflanzlichen Geweben. *Jahrb. f. wiss. Bot.*, Bd. XXX.

1900. DAWSON, MARIA. Further Observations on the Nature and Functions of the Nodules of Leguminous Plants. *Phil. Trans. Roy. Soc.*

1896. CZAPEK, F. Zur Lehre von den Wurzelausscheidungen. *Jahrb. f. wiss. Botanik*, Bd. XXIX.

1892. HEDLUND, T. Kritische Bemerkungen über einige Arten der Flechtengattungen Lecanora, Lecidea, und Micarea. *Bihang till K. Sven. Vet.-Akad.* Handl., Bd. XVIII.

1899. PEIRCE, G. J. The Nature of the Association of Alga and Fungus in Lichens. *Proc. Cal. Acad. Sci., 3d Ser.* (Bot.), Vol. I, No. 7.

1894. SPALDING, V. M. The Traumatropic Curvature of Roots. *Ann. Botany*, Vol. VIII.

1893. PEIRCE, G. J. Structure of the Haustoria of Some Phanerogamic Parasites. *Ann. Botany*, Vol. VII.

1897. FISCHER, A. Vorlesungen über Bakterien. Jena

1901. SMITH, E. F. Entgegnung auf Alfred Fischer's "Antwort" in betreff der Existence von durch Bakterien verursachten Pflanzenkrankheiten. 2ter Theil. *Centralbl. f. Bakteriol., Parasitenkunde u. infektionskrankh.*, II Abtheilung. (The special literature is here cited.)

1894. PEIRCE, G. J. A Contribution to the Physiology of the Genus Cuscuta. *Ann. Botany*, Vol. VIII.

1901. CANNON, W. A. The Anatomy of Phoradendron villosum Nutt. *Bull. Torr. Bot. Club*, Vol. XXVII.

1893. SCHNEIDER, A. The Morphology of Root-tubercles of Leguminosæ. *American Naturalist*, Vol. XXVII.

1901. LIFE, A. C. The Tuber-like Rootlets of Cycas revoluta. *Bot. Gazette*, Vol. 31.

1892. PFEFFER, W. Ueber Anwendung des Gipsverbandes für pflanzenphysiologische Studien. *Ber. d. K. Sächs. Gess. d. Wiss.*

1894. NEWCOMBE, F. C. The Influence of Mechanical Resistance on the Development and Life-period of Cells. *Bot. Gazette*, Vol. XIX.

1894. RICHTER, J. Ueber Reaktionen der Characeen auf äussere Reize. *Flora*, Bd. LXXVIII.

1897. MAZÉ. Fixation de l'azote libre par la bacille des nodosités Légumineuses. *Ann. de l'Inst. Pasteur*, T. x 1.

1900. MAGNUS, W. Studien an den endotrophen Mycorrhiza von Neottia Nidus avis L. *Jahrb. f. wiss. Bot.*, Bd. XXXV.

EXPLANATION OF PLATE XXIX.

Most of the figures were drawn with the aid of a Leitz drawing prism ocular, the rest with an Abbé camera lucida.

Fig. 1. Two root-hairs of Bur Clover infected by tubercle bacteria, showing the characteristic bending at the point of infection; x 50.

Fig. 2. The lower of the two root-hairs of fig. 1. The mass of bacteria in the concavity of the coil and the infection thread running from this point through the hair; x 300.

Fig. 3. Another infected and coiled root-hair with the infection thread growing close to the nucleus of the hair; x 300.

Fig 4. A section showing the course of the infection thread from the base of the hair *(r. h.)* to the layer of cells which gives rise to the tubercle, the same layer which, under other conditions, would give rise to a lateral root; x 360.

Fig. 5. A section showing a young tubercle (left) and a young lateral root (right) developing from the same layer; x 20.

Fig. 6. The same section enlarged. In the lateral root (right) differentiation of tissues is already taking place, but not in the tubercle (left): x 360.

Fig. 7. Longitudinal section of a tubercle, young and still growing. The largest part of the tubercle is composed of bacteria-containing cells. Outside of this mass is the comparatively thin layer in which are the small and scattered vascular bundles. Beyond this is the protective tissue, powdery on the outside, which is continuous with the cortex of the root. At *a–b* is the tubercle meristem, which forms daughter-cells both forward and backward, as does the growing-point of a root; x 35.

Fig. 8. A typical infected tubercle cell, thin-walled, with the cytoplasm surrounding a large single vacuole, and the nucleus reduced to a small lumpy plate. Cytoplasm stained with orange G., but the color obscured and dulled by the purple of the bacteria stained by anilin gentian violet. The nucleus is purplish from the mixture of anilin safranin and anilin gentian violet, a wholly different color from that of a healthy nucleus stained and differentiated by Flemming's triple stain and Gramm's iodine solution. Intercellular spaces are shown; x 660.

Fig. 9. Tubercle cell containing only small number of bacteria (at *B*) and enclosing some starch-grains *(S)*. The nucleus has degenerated only somewhat and the vacuolization of the cytoplasm has not progressed far; x 820.

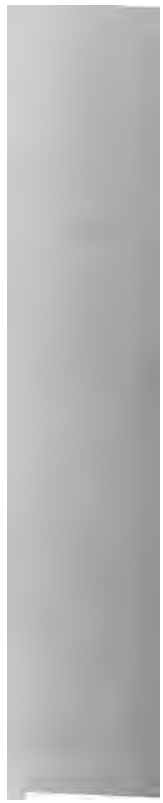
Fig. 10. Section of a tubercle, part of tubercle shown in fig. 7, near the meristem. The direction in which the meristem lies is indicated by the arrow at the side. The section was stained by Flemming's triple stain and differentiated, after the anilin gentian violet, by Gramm's iodine. Thus the infection threads are clearly brought out. Note the course of the infection threads, definitely toward the tubercle meristem and generally toward the nuclei of the cells entered; x 200.

Fig. 11. One cell from fig. 10 (nearly in the center of the figure) showing the solid infection strand (zoöglœa) in which the separate bacteria can be distinguished; x 1000.

Fig. 12 and 13. Tubercle cells showing the infection threads growing in definite direction, generally toward the nuclei of the cells; x 820.

Fig. 14, *a–d*. A series of cells from the meristem backward in a longitudinal section of a tubercle. The section is stained by Flemming's triple stain, but not differentiated by Gramm's iodine, hence infection threads do not show. In fig. 14 *a*, two meristem cells show at *x–y*, with daughter-cells both forward and backward. *Z* = already infected and degenerating cell. In 14 *b*, degeneration of the cell and especially of the nucleus is shown by cells *c*, *i*, *h*. Degeneration of the nucleus, obscuring of the color of the cytoplasm by the many bacteria in it, and the formation of the characteristic vacuole shows in 14 *c*, especially the two upper cells. Fig. 14 *d* shows the same still more markedly, especially the uppermost cell. In this series the effect of bacterial infection of the cells is clearly exhibited; 14 *a*, x 270; 14 *b–d*, x 300.

Fig. 15. Division of an already, though recently, infected daughter-cell of the tubercle meristem; x 300.



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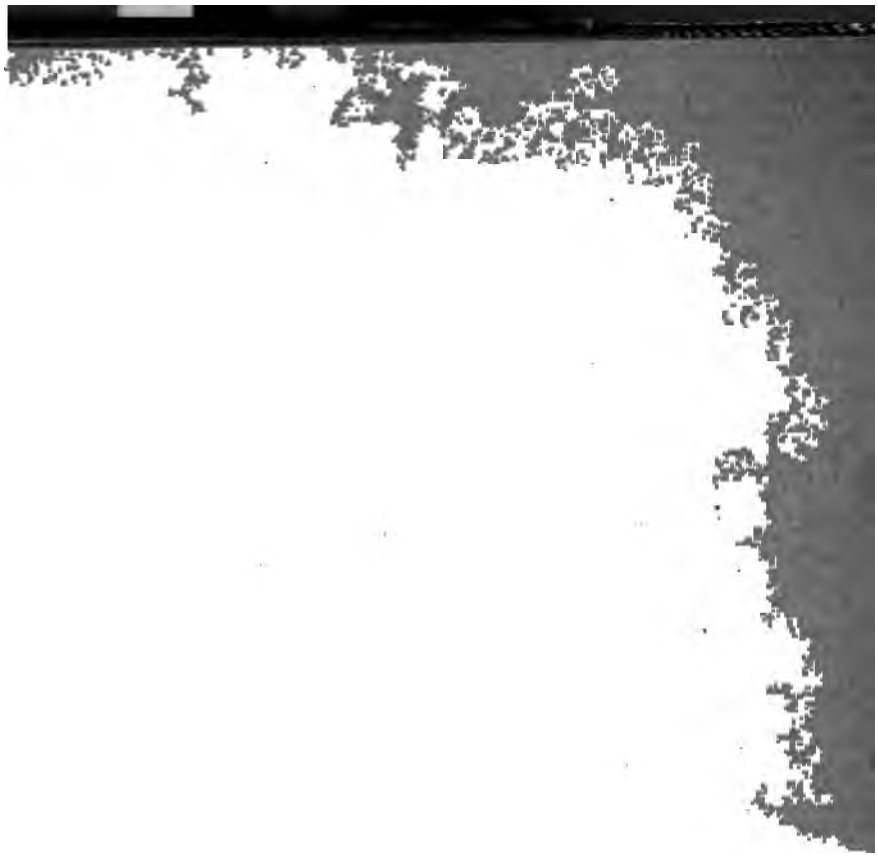

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Spindle Formation in the Pollen-Mother-Cells of Cassia tomentosa L

BY
HENRI T. A. HUS

WITH THREE PLATES

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SPINDLE FORMATION IN THE POLLEN-MOTHER-CELLS OF CASSIA TOMENTOSA L.

BY HENRI T. A. HUS.

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INTRODUCTION.

Cassia tomentosa L. possesses a number of qualities which particularly recommend this plant to the cytologist. Chief among these are the large number of buds produced and the protracted flowering period. The stages of cell-development which are of greatest interest at the present time are passed through very rapidly, and are also the most difficult to fix; hence the collection of an immense amount of material is necessary to ultimate success. At the same time the protracted flowering period affords an opportunity to determine at leisure which is the best fixing-medium and what shall be the duration of the fixing-period.

The following fixing-fluids were tried:

1. Strong Flemming:
   1 per cent. chromic acid 15 pts.
   2 per cent. osmic acid 4 pts.
   Glacial acetic acid 1 pt.
2-6. No. 1 diluted with 1-5 volumes of water.
7. 1 per cent. chromic acid 15 pts.
   2 per cent. osmic acid 4 pts.
   1 per cent. glacial acetic acid 1 pt.
8-12. No. 7 diluted with 1-5 volumes of water.
13. 1 per cent. chromic acid 15 pts.
   2 per cent. osmic acid 4 pts.
14-18. No. 13 diluted with 1-5 volumes of water.
19. ½ per cent. chromic acid 15 pts.
   2 per cent. osmic acid 4 pts.
   Glacial acetic acid 1 pt.
20. No. 19 diluted with 1 volume of water.
21. 1 per cent. chromic acid.
22. 2 per cent. osmic acid.
23. 1 per cent. osmic acid.
24-26. 1, 5 and 10 per cent. aqueous solution of bichromate of potash.
27. Tellyesniczky's fluid (Arch. mikr. Anat. 52:247. 1898):
   Bichromate of potash 3 gm.
   Glacial acetic acid 5 cc.
   Distilled water 100 cc.
28-32. No. 27 diluted with 1-5 volumes of water.
33. Müller's solution:
   Bichromate of potash 2-2½ pts.
   Sulphate of soda 1 pt.
   Distilled water 100 pts.
34. Zenker's mixture:
   Corrosive sublimate 5 per cent.
   Glacial acetic acid 5 per cent.
   Müller's solution 90 per cent.
35. [1] Saturated aqueous solution of bromine 1 pt.

[1] The material was immersed for two hours in this solution, and then removed to Flemming's strong mixture, where it remained eight hours.

	Distilled water	9 pts.
36.	⅓ per cent. iodine 1⅓ per cent. potassium iodide Distilled water	1 pt.
	Distilled water	9 pts.

I cannot here enter upon an extensive discussion of the effects of these fixing-fluids. It is sufficient to say that material fixed for ten to twelve hours in Flemming's strong mixture showed as a rule no shrinkage; while material fixed in other solutions (especially in the very dilute ones) or left longer than twelve hours in Flemming's strong mixture, showed considerable shrinkage, with the exception perhaps of that fixed with Tellyesniczky's fluid, which gave fairly good results.

In fixing, two rules were observed: one, always to fix in the field, since only in material fixed in this manner were some of the earlier stages found in any number; the other, to keep the material submerged, for only then could the necessary penetration of the fixing-fluid take place. Frequently air-bubbles adhered to the anthers, especially if the latter were not separated, causing them to float. To prevent this a very simple device was used: a disc was cut off the lower end of the cork of the neckless fixing-bottle and attached to a glass rod passing through the cork; this kept the material submerged, while a notch in the edge of the disc allowed the escape of any air-bubbles.

After fixing, the material was washed from six to eight hours in running water; a longer stay in water apparently injured the finer structure of the cytoplasm.

At this stage the material was sorted and then dehydrated. It appeared here that a prolonged stay in weak alcohol produced shrinkage. After reaching 95 per cent. alcohol, the material remained for periods of about twelve hours each, in absolute alcohol, a mixture of equal parts of absolute alcohol and bergamot oil, bergamot oil, a mixture of equal parts of bergamot oil and 43° C paraffin, 43° C paraffin and 54° C paraffin. It was then imbedded in 71° C

paraffin, the use of which was found necessary to obtain good ribbons. Sections 3-5 μ. thick were cut and fixed on the slide by the water-albumen method.

Flemming's triple stain was used, with some modifications. The length of time for each stain was ascertained separately. It was finally found advisable not to use the safranin solution usually employed, but to substitute for it Babes' safranin A, consisting of a mixture of equal parts of a saturated alcoholic solution of safranin and a saturated aqueous solution of safranin, because the former had a tendency, when old, to give a muddy appearance to the cytoplasm.

In the case of orange G, even weak solutions took out too much blue. By substituting for this a solution of iodine and potassium iodide of the same strength and composition as the fixing-fluid mentioned under number 36, much better results were obtained. In fact the potassium iodide iodine seemed to fix the blue in the fibers, so that absolute alcohol and clove-oil could be relied upon to wash out the superfluous blue.

The staining process was as follows: After dissolving the paraffin in xylol and removing all traces of the latter by a double washing in 95 per cent. alcohol, the slides were placed in the safranin for five minutes and then decolorized in absolute alcohol to which 0.1 per cent. hydrochloric acid had been added. After washing thoroughly in water to remove all traces of the acid, the slides were placed for exactly five minutes in a concentrated aqueous solution of gentian violet, and then washed off in water; after which they were immersed for twenty seconds in the solution of iodine and potassium iodide above referred to. This appeared to fix the blue in an admirable way, so that after dehydrating in absolute alcohol for one second, the preparations, in clove-oil, could be watched under the microscope until the cytoplasm was perfectly clear, the fibers a dark blue, and the granular zone brown-violet. The nucleolus was a bright red as well as the chromatin, except in the earlier stages, when the chromatin stained blue.

DESCRIPTION.

The nuclei of the pollen-mother-cells of *Cassia tomentosa*, in the stage indicated in figure 1, show a nucleolus, which is sometimes vacuolate, and a broken up chromatin thread. The former stains red, the latter blue or red. The cytoplasm is composed of two parts, one apparently fibrous,[1] the other granular. The first is composed of thinner or thicker fibers running in every direction, but more especially from the nuclear wall to the cell-wall, so that the meshes which they form are elongated in a radial direction (fig. 1). The meshes are smallest in the neighborhood of the nucleus, and gradually increase in size toward the periphery.

The other component of the cytoplasm appears in the form of larger or smaller granules situated between and upon the fibers, and showing a tendency to accumulate in larger or smaller masses, especially at the intersection of the fibers. The fibers can be stained blue with gentian violet, while the granules stain brown-violet. The whole presents a very uniform appearance, except here and there where a thicker fiber or a denser accumulation of granules shows more prominently.

Scattered irregularly through the cell may be seen small dark round bodies, apparently oil globules, to judge by their appearance in fresh material. It may be of interest to note that while these bodies were very numerous in some cases, especially in the early stages, in others they were rare and were then found but seldom in the later stages.

Gradually the cytoplasm around the nucleus begins to change in appearance: the change takes place in a single layer of meshes immediately adjacent to the nuclear wall; these meshes become smaller, due perhaps to the interpolation of new fibers, and to the appearance immediately around the nucleus of granular matter, situated upon and between the fibers, often completely obscuring them. The meshes now become elongated parallel to the nuclear wall;

[1] The description of the structure of the cytoplasm refers to microscopical appearance only, without indicating whether it is a reticulum or a foam-structure.

they become very narrow, being usually from two to three times as long as broad. The fibers are covered with granules; they begin to stain a deeper blue than the other fibers. This appearance extends only partially around the nucleus, usually about half way (fig. 2.)

Changes now begin to take place in the cytoplasm farther away from the nucleus: the cytoplasm loses its uniform appearance; here and there fibers have begun to thicken, or granules have accumulated upon them. These fibers stain a deep violet and show sharply against the lighter background (fig. 3). They are arranged in more or less conical groups with their bases directed toward the nucleus.

Very gradually, almost imperceptibly, the granules around the nuclear wall increase in number, and a second layer of elongated meshes is laid down next the one already formed. In the meantime the granules situated upon and between the fibers have disappeared, and the fibers have become thick and smooth (fig. 3). We now see the nucleus partly surrounded by a number of rectangular meshes elongated in a direction parallel to the nuclear wall.

These processes seem to continue for some time, until finally three or four layers of elongated meshes have been laid down around the nucleus. Then the radial fibers of these meshes begin to disappear, leaving a zone composed of fibers running parallel to the nuclear wall. That this zone is not attached closely to the latter can be easily observed in later stages when one or more spaces exist between the nuclear wall and the fibrous felt-like zone, which stands out clearly as a dark blue band.

In the meantime granules have continued to accumulate around the nucleus, and the meshes of the cytoplasm adjoining the felt-like zone have grown smaller. Together they form a distinct zone (the granular zone) which possesses a distinct outer boundary.

The cell now presents the appearance indicated in figure 4. The chromatin appears in the form of lumps, which like the nucleolus, stain a brilliant red. Here and there linin, in the form of violet-staining granular threads, may

be seen connected with the chromosomes. The nucleus is more or less completely surrounded by a zone of smooth parallel fibers which stain a dark blue. Next these lies the granular zone, occupying about half the remaining cell-space, and composed of a reticulum of small, more or less isodiametric meshes, between which (especially at the intersection of the threads) a great number of larger and smaller granules may be seen. Surrounding this zone (which stains brown-violet) is a dark blue line apparently composed of fibers upon which granules, large and small, are situated. These fibers are not continuous, and the number of granules upon them is not the same in all places, so the ring which they form is an irregular one. It stains a deep blue, very effectually marking the boundary between the brown-violet granular zone and the outer gray-yellow cytoplasm, which is composed of fibers forming large irregular meshes. Upon and between these fibers are larger or smaller masses of granules. The granular zone does not present an absolutely uniform appearance; in places fibers upon which granules have accumulated stand out clearer than others. Usually they run parallel to, or at a small angle with the nuclear wall. At first only one or two are seen, but they rapidly increase in number. Soon those nearest the nuclear wall seem to establish connection with some of the fibers of the felt-like zone (fig. 5); gradually other, deeply-staining fibers are added, all running at a greater or lesser angle to the nuclear wall, and finally arranging themselves in conical groups with their bases resting on the fibrous zone. Not only one cone like this may be observed, but several (fig. 6). During this time the number of prominent fibers in the granular zone continues to increase. Frequently a space can be observed, at least in places, between the nuclear wall and the felt-like zone.

At about this stage the nucleolus begins to disappear; in the preceding one, however, the nucleolus was still present, and the chromatin was in the form of curved, rod-shaped chromosomes, or else in the form of rings, formerly considered so typical of the Liliaceæ.

The linin of the nucleus has gradually become more prominent, staining a darker blue; but while in the preceding stage it showed as fine granular threads, it now (fig. 6) appears as smooth fibers, running through the nucleus, and usually more or less parallel to each other and to the axis of the largest cone. The fibers stain blue.

The cones, which are composed of granular threads, increase in number. Some are larger; some smaller. Usually one has the ascendency over the others, and this, as a rule, is the first cone formed; it appears ordinarily at one end of the ellipsoidal nucleus. As has been said, it is usually parallel to the axis of this largest cone, and consequently more or less parallel to the longer axis of the nucleus, that the linin threads are directed. This seems to indicate a polarity of the cell.

In figure 6 we find the cones composed of granular threads; these threads are in connection with the fibers of the felt-like zone. In the next stage we find cones, large and small, composed of smooth fibers. These fibers apparently originate from the felt-like zone. But do they push out of their own accord? or are they pulled out by the granular fibers? or are the cones a result of a combination of the granular fibers and those of the felt-like zone? Judging from my preparations the last seems to be the most probable.

As soon as the fibers of the cone have become smooth, the nuclear wall just below the cone breaks down, and the fibers penetrate into the nuclear cavity, where they anastomose with the linin threads. The two kinds of fibers can hardly be distinguished, for while it is true that the fibers originating from the cytoplasm are thicker than those formed from the linin, even this difference very soon disappears.

Even at this early stage the fibers arrange themselves in bundles (fig. 7), an arrangement which is far less prominent in the fully developed multipolar stage, but which reappears in the bipolar spindle.

The chromosomes retain their original position for a long

time after the nuclear wall has disappeared. Only when the multipolar spindle has been fully formed, and in fact not until a rearrangement of the poles (with a view to the formation of a bipolar spindle) has begun, do they begin their migration toward the spot where the equatorial plate is to be formed. Throughout the formation of the cones and the subsequent changes from multipolar to bipolar spindle, no indication of a centrosome could be observed.

As soon as the fibers composing the cones have become smooth, the nuclear wall breaks down and the blue-staining fibers penetrate the nuclear cavity, where they lie in contact with the bright red chromosomes. The multipolar spindle thus formed has a variable number of cones; the largest number observed in a single section was eight (fig. 10); but as the cell was always cut into a number of sections, the number of cones must be greater.

The granular zone now presents a uniform appearance, and stains from yellow-brown to brown-violet; the granular threads which were formerly so prominent have disappeared; nothing remains but small meshes which are fairly uniform in size. Between and upon the threads composing the meshes a large number of granules can be observed. The poles protrude into the granular zone. Surrounding the granular zone we find an irregular line still staining a deep blue. The cytoplasm outside this line retains the same structure as in the last stage.

In the formation of the multipolar spindle a highly interesting phenomenon was observed. It has been stated before that, as a rule, a cone situated at one end of the nucleus had from the first an ascendency over the others; and while it was not observed that another cone (situated directly opposite this one) was always the second to be formed, yet one could not fail to see that after a little while two cones (one at each end of the nucleus) were more prominent than the others, or were even the only cones present, in which case the nuclear wall disappeared beneath these cones while persisting everywhere else. Such a case is shown in figure 8. In nearly every instance this promi-

nence of two cones could be noted. If my observations are correct, we have in *Cassia tomentosa* a most interesting transition stage between the multipolar polyarchal and the multipolar diarchal spindle.

The largest number of poles of the multipolar spindle observed in cross section was eight; the most frequent number noted in a section was from four to six. In what manner the multipolar spindle finally became a bipolar one could not be determined with certainty; the cones approached each other in two groups (fig. 12), finally forming the bipolar spindle.

The cell now presents the appearance indicated in figure 13. The chromosomes lie in the equatorial plate. In polar view, twelve chromosomes can be observed. The spindle has sharp-pointed poles. Apparently there are here three kinds of fibers; some having the appearance of strands are attached to the chromosomes; some run from pole to pole; while from each pole mantle-fibers diverge into the granular zone. The fibers stain a deep blue; the granular zone is yellow-brown; but the irregular line around the granular zone is as blue and sharp as it was before. The poles of the spindle project into the granular zone.

The wandering of the chromosomes toward the poles now begins in the usual manner; the continuous fibers still run from pole to pole; while the mantle-fibers become more prominent.

At this stage we find, sometimes inside the granular zone, sometimes outside it, small round bodies which stain red, sharply contrasting with other similar bodies, lying on the outskirts of the granular zone, which stain a deep violet, and look like the oil-globules previously observed.

Finally the chromosomes reach the poles, and the continuous fibers as well as the mantle-fibers assume a wavy appearance. The mantle-fibers during the last stage diverge more than formerly.[1] Why the continuous fibers should assume this wavy appearance is not clear. Certainly the

[1] Perhaps it would be well to state here that these mantle-fibers were not equally prominent in all preparations. In fact in some cases no trace of them could be found.

suggestion of Williams (1899, p. 194; *vide* Went, 1887, p. 258, figs. 8-10) that it is caused by the drawing together of the poles, appears very acceptable. In the case of *Cassia tomentosa* the spindle poles either reach to the outer edge of the granular zone, or else pass through it (figs. 13, 14), yet the daughter nuclei are always formed well within it.

The most interesting point at this stage is the behavior of the granular zone, which shows a tendency to accumulate around the daughter nuclei as they are about to be formed. Gradually it surrounds them on all sides except where the connecting fibers remain. As soon as a wall is formed around the daughter nuclei, they become entirely surrounded by granular matter.

It was the last stages of this process which were particularly easy to follow in my preparations. To say that the granular matter encroached upon that part of the cell which contained the yet remaining continuous fibers would not be quite true; the fibers seemed rather to thicken at various places in the immediate neighborhood of the daughter nuclei, and this apparently at the cost of the immediately adjacent parts of the fibers. This process continued until nothing was left of that part of the continuous fibers but a granular thread, which sooner or later lost its continuity, so that gradually the place where the threads were, was entirely taken up by granules.

This process takes place very gradually, beginning at the sides of the daughter nuclei and proceeding toward the axis of the cell. At first the meshes formed by the transformation of the connecting fibers into granular matter are fairly large, but soon they can no longer be differentiated from those of the rest of the granular zone. This change, especially in its later stages, is very slow, and may not be consummated until the daughter nuclei are already forming spindles.

The daughter nuclei now lie in a granular mass in no way different apparently from that which surrounded the mother nucleus. The shape of the granular zone is not quite circular, but more or less ellipsoidal, slightly constricted about the middle.

The shape of the daughter nuclei is decidedly ellipsoidal, with the longest axis at right angles to the main axis of the previous spindle. The daughter chromosomes are irregular in shape: sometimes they take the form of lumps; at other times they appear as chains of granules. They stain red and are connected by bands of finely granular matter. A body which seems to be a nucleolus appears in each daughter nucleus.

Owing to the presence of a dense granular zone around each daughter nucleus, it was impossible to follow in this case the stages just preceding the formation of the multipolar spindle; yet what could be seen was sufficient to indicate that these stages agreed in the main with the corresponding stages of the mother nucleus. The differences observed are to be explained by the shape of the spindle, which in all cases is of a pronounced multipolar diarchal type.

The first indication of spindle formation that could be observed was the appearance of a mass of clean-cut, closely-woven fibers surrounding each daughter nucleus. These fibers were particularly distinct where the nuclear wall had shrunken somewhat. They stained a deep blue (fig. 17).

The multipolar spindles of the daughter nuclei are formed in a manner similar to that of the mother nucleus. Linin fibers become more and more prominent within the daughter nucleus; they run more or less parallel to the longest axis of the nucleus in which they are formed. Cones begin to appear here and there on the nuclear wall, usually at the ends; they increase in number. In some places the nuclear wall breaks down; the cytoplasmic fibers enter the nuclear cavity, and become merged with the threads of nuclear origin. Finally a multipolar spindle is formed, which, as has been previously stated, is of a pronounced multipolar diarchal type. The narrow, compressed appearance of the entire multipolar figure is here very striking, far more so than in the multipolar spindle of the mother nucleus. In no case was as large a number of poles observed as in the

corresponding stage of the mother nucleus. The planes in which the spindles of the daughter nuclei lie are sometimes at right angles, sometimes parallel to each other.

Finally the daughter chromosomes reach the poles and become surrounded with a membrane. The chromatin is in the form of granules, arranged in some places to form a thread, and interconnected by bands of the same finely granular substance which we saw connecting the chromosomes of the daughter nuclei. The nucleolus, which disappeared at the time of the dissolution of the membrane of the daughter nuclei, now again makes its appearance.

The granular zone surrounds each of the four daughter nuclei very much as in the stages just following the first division of the mother nucleus; but instead of filling up the entire space between the nuclei, it merely surrounds them, leaving a clear space in the middle through which fibers may be seen running from one nucleus to the other.

DISCUSSION.

In the formation, development, and subsequent behavior of the spindle there are two points of special interest; the nature of the multipolar spindle, and the origin of the cones.

THE NATURE OF THE MULTIPOLAR SPINDLE.

In the first as well as in the second divisions of the pollen-mother-cells of *Cassia tomentosa* there is more or less approach to the spindle formation described for vegetative cells. Instead of a multipolar polyarchal spindle Anlage (Strasburger, 1900, p. 121) such as we usually meet with in pollen-mother-cells, we have here a more or less multipolar diarchal spindle. At the very beginning we meet with an indication of this. The nucleus is usually ellipsoidal. The first cone (afterwards the largest cone) appears ordinarily at one end of the ellipsoidal nucleus. The second cone formed lies in many instances directly opposite the first one, or in other cases, the cone which

occupies this position becomes, as a rule, more prominent than those surrounding it. These two cones remain the most prominent ones. More or less parallel to the axis of these cones, and consequently more or less parallel to the longer axis of the nucleus, run the linin threads. The multipolar diarchal nature of the spindle is even more prominent in the second division.

Thanks to the numerous investigations of recent years, we are now able to compare a number of modes of spindle formation. Strasburger (1900, p. 118) distinguishes three types of spindle formation; the multipolar polyarchal, the multipolar diarchal, and, opposed to these, the bipolar diarchal.

Multipolar polyarchal spindles are characteristic of the first division of pollen-mother-cells *(Lilium, Passiflora, Lavatera, Cobæa, Gladiolus)* and of spore- and embryosac-mother-cells (Osterhout, 1897; Strasburger, 1900, p. 121). In *Cassia tomentosa* we have a multipolar diarchal spindle Anlage in the first division, which thus forms a connecting link between the multipolar polyarchal spindle Anlage of reproductive cells and the multipolar diarchal spindle described for vegetative cells, i. e., the root-tips of *Vicia* (Strasburger, 1900, p. 116), of *Allium* (Nemec, 1899*a*), and of ferns (Hof, 1898, p. 169). In *Nymphæa alba* (Strasburger, 1900, p. 122) the spindle Anlage of the mother nucleus does not differ essentially from that in the daughter nuclei described by Strasburger for *Iris squalens*, where it is multipolar diarchal.

It seems therefore that there exists no sharply marked difference between spindle formation in reproductive and in vegetative cells, and that the two processes are closely related.

As suggested by Strasburger (1900, p. 122), the various modes of spindle formation would readily lead one to suppose them influenced by polarity: "Eine Pollen- und Sporenmutterzelle könnte auch in ihren Protoplasten multipolar sein und so die polyarche Anlage der Kernspindel bedingen. Auch liesse sich denken, das die

diarche Anlage der Kernspindeln in Gewebezellen eine Folge ihrer longitudinalen und radialen Polarität sei."

Polarity is in many cases probably determined by causes outside the cell; for though in the first division of the pollen-mother-cells and in most divisions in vegetative cells, the long axis of the spindle coincides with the long axis of the cytoplasmic mass, yet there are numerous cases (both in reproductive and in vegetative cells) where the long axis of the spindle is placed at right angles to the long axis of the cell.

Instances of these are the spindles of the daughter nuclei in pollen-mother-cells, where the nuclei have probably a reciprocal influence. Though the axes of the two spindles are generally parallel and at right angles to the longest axis of the cell, yet it sometimes occurs that the two spindles lie in planes which are at right angles to each other. More striking are those divisions in the cambium cells where the axis of the spindle lies at right angles to the long axis of the cell.

It seems probable that we must in many cases look outside the cell for the cause of the polarity, as is especially indicated by the observations of Nemec (*Flora*, 1899*b*, p. 219) on cell-division in potato-tubers, where, in a cell (the surrounding cells having died) the spindle formation did not take place in "monaxial" fashion, but cones were formed on all sides.

THE ORIGIN OF THE CONES OF THE MULTIPOLAR SPINDLE.

But few cytologists have investigated closely the origin of the cones of the multipolar spindle. The first to devote his special attention to this subject was Belajeff (1894), in whose paper "Zur Kenntniss der Karyokinese bei den Pflanzen" were published the results yielded by his observations on the division of the pollen-mother-cells of various species of *Larix*. Since then, in addition to Strasburger's observations, the spore-mother-cells of *Equisetum* (Osterhout, 1897), and the pollen-mother-cells of *Cobæa* (Lawson,

1898), *Passiflora* (Williams, 1898), *Lilium* (Grégoire, 1899), *Lavatera* (Byxbee, 1900), *Gladiolus* (Lawson, 1900), and now *Cassia*, have been studied more particularly in this regard. The results of these studies are a number of observations which agree in the main, though in minor details they frequently differ.

It may therefore be of interest to compare the various modes of formation and development of the cones of the multipolar spindle in the spore- or pollen-mother-cells of these plants.

About even the earliest stages the observations frequently differ. In *Larix*, *Cobæa*, and *Cassia* the first stages observed are described as showing a more or less radial arrangement of the meshes of the cytoplasm (in *Cobæa* in the immediate neighborhood of the nucleus only). Similar observations have been made on the pollen-mother-cells of *Lilium speciosum* (Grégoire, 1899), and by myself on those of *Lilium Humboldii*. In *Equisetum* and *Gladiolus* this radial arrangement is not specially mentioned; but in regard to *Passiflora* and *Lavatera* it is distinctly stated that this radial arrangement of the cytoplasmic meshes does not appear until an elongation of the meshes parallel to the nuclear wall has taken place. In all plants mentioned (with the exception of *Cobæa*) this parallel elongation of the cytoplasmic fibers has been observed and followed with great care, especially in *Lavatera*. Here the manner of the formation of the meshes is identical with that observed in *Cassia*, except perhaps, that in *Cassia* the fibrous zone in its first stages is not so complete. But sooner or later the number of rows of parallel elongated meshes increases to such an extent that the felt-like mass is easily discernible. At this stage it is figured by a number of authors. Strasburger (1888) figured and mentioned it; but Belajeff (1894) was the first to describe this felt-like zone: "Die erste Veränderung, welche ich bei in Theilung begriffenen Zellen habe beobachten können, war die Bildung einer den Kern umhüllenden, dichten, filzartigen Schicht, welche auf den ersten Blick als eine concentrisch um den Kern gewun-

denener Fadenknauel erscheint. Eine nähere Untersuchung feinster Schnitte zeigt jedoch, dass diese Filzschicht aus der Kernwandung parallel in die Länge gezogenen Schlingen (Maschen) besteht."

This description would fit the felt-like zone occurring in the pollen-mother-cells of all the plants just mentioned; but Belajeff's words, "Die obenbeschrieben Umgruppirung der Plasmafäden ist, wie es scheint, die Folge ihrer Contraction und des Zusammenziehens der Schlingen (Maschen) um den Zellkern," do not agree with the views of others in regard to this matter. Both Byxbee and Williams are of the opinion that the meshes become elongated because of a drawing-out. This then would be caused, not by a contraction of cytoplasmic threads, but by their expansion. What causes this expansion is not yet explained, but an increase in size of the nucleus is not improbable, and however slight, would produce the appearance described. Observations on *Cassia* tend to confirm this supposition. But whatever the manner of its origin, a weft-like zone seems to be formed around the nucleus in nearly all cases.

The manner in which the fibers of cytoplasmic origin form cones seems to differ in the various plants observed. In this connection it is necessary to call attention to the fibers which may be seen in the cytoplasm at the time of the formation of the fibrous zone. In *Cassia* these fibers appear when the first changes in the cytoplasm immediately adjoining the nucleus begin to take place. At various points the presence of granular fibers, which stain a deep violet and which are sometimes arranged in conical groups, becomes apparent. They remind one of the "irregular, deeply staining strands" of *Passiflora* (Williams, 1899, p. 191) or of the cones of *Equisetum* (Osterhout, 1897, p. 161, fig. 4). In *Cassia* these fibers apparently establish a connection with the fibers of the fibrous zone, which latter gradually become parallel, or nearly so, to the thicker granular fibers running at a greater or lesser angle to the nuclear wall. The origin of the cones is apparently determined by the thicker granular fibers. No drawing-out of the cones takes place.

[redacted]

c. The fibers forming elongated meshes around the nucleus become smooth (fig. 3).

d. A felt-like zone is formed partially or completely surrounding the nucleus; granular linin threads appear within the nucleus; the granular zone now takes up about one-half the remaining space (fig. 4.)

e. The deeply-staining rough fibers of the cytoplasm, united into cones, establish connection with those of the felt-like zone (fig. 5).

f. The linin threads of the nucleus become more prominent and finally smooth. They run parallel to each other and to the axis of the cone which has the ascendency over the others.

g. As soon as the threads of a cone become entirely smooth, the nuclear wall breaks down at the base and the linin and kinoplasmic fibers anastomose. The fibers become grouped into bundles (fig. 7).

h. A multipolar spindle is formed, two cones of which, situated opposite each other, are more prominent than the rest. Sometimes but two cones are present (fig. 8). The two prominent cones finally absorb the others, thus forming a bipolar spindle.

3. The spindle formation for the second division takes place in the manner described for the first, but is even more pronouncedly multipolar diarchal.

4. The spindles of the daughter nuclei sometimes lie in planes which are sometimes at right angles, sometimes parallel to each other.

5. The spindle formation in *Cassia tomentosa* L. forms a connecting link between the multipolar polyarchal spindle Anlage ordinarily met with in dividing pollen-, spore-, and embryosac-mother-cells, and the multipolar diarchal spindle-Anlage described for vegetative cells.

In conclusion I beg to acknowledge my great indebted-

ness to Dr. W. J. V. Osterhout, at whose suggestion the work was undertaken, and under whose direction it was for the greater part carried out.

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BIBLIOGRAPHY.

1894. Belajeff, W. Zur Kenntniss der Karyokinese bei den Pflanzen. *Flora*, Bd. LXXIX, p. 430.

1900. Byxbee, E. S. The Development of the Karyokinetic Spindle in the Pollen-Mother-Cells of Lavatera. *Proc. Calif. Acad. Sci.*, 3d Ser. (Bot.), Vol. II, No. 2.

1899. Grégoire, V. Les cinèses polliniques chez les Liliacées. *La Cellule*, T. XVI, 2e fasc., p. 235.

1898. Hof, A. C. Histologische Studien an Vegetationspunkten. *Bot. Centralblatt*, Bd. LXXVI, p. 65.

1897. Juel, H. O. Die Kerntheilungen in den Pollenmutterzellen von Hemerocallis fulva, und die bei denselben auftretenden Unregelmässigkeiten. *Jahrb. f. wiss. Bot.*, Bd. XXX, p. 205.

1898. Lawson, A. A. Some Observations on the Development of the Karyokinetic Spindle in the Pollen-Mother-Cells of Cobæa scandens Cav. *Proc. Calif. Acad. Sci.*, 3d Ser. (Bot.), Vol. I, p. 169.

1900. ———Origin of the Cones of the Multipolar Spindle in Gladiolus. *Bot. Gazette*, Vol. XXX, p. 145.

1897*a*. Mottier, D. M. Beiträge zur Kenntniss der Kerntheilung in den Pollenmutterzellen einiger Dicotylen und Monocotylen. *Jahrb. f. wiss. Bot.*, Bd. XXX, p. 169.

1897*b*. ———Ueber das Verhalten der Kerne bei die Entwickelung des Embryosäcks und die Vorgänge bei der Befruchtung. *Jahrb. f. wiss. Bot.*, Bd. XXXI.

1899*a*. Nemec, B. Ueber die karyokinetischen Kerntheilung in der Wurzelspitze von Allium Cepa. *Jahrb. f. wiss. Bot.*, Bd. XXXIII, p. 313.

1899*b*. ———Ueber Kern und Zelltheilung bei Solanum tuberosum. *Flora*, Bd. LXXXVI, p. 214.

1897. Osterhout, W. J. V. Ueber Entstehung der karyokinetischen Spindel bei Equisetum. *Jahrb. f. wiss. Bot.*, Bd. XXX, p. 159.

1896. Rosen, F. Beiträge zur Kenntniss der Pflanzenzelle. *Cohn's Beiträge zur Biologie der Pflanzen*, Bd. VII, p. 249.

1888. Strasburger, E. Ueber Kern und Zelltheilung. Jena.

1900. ———Ueber Reduktionstheilung, Spindelbildung, Centrosomen und Cilienbildner im Pflanzenreich. Jena.

1884. Tangl. Zur Lehre von der Kontinuität des Protoplasma. *Sitzber. d. k. k. Akad. d. Wiss.*, Bd. XC.

1887. Went, F. A. F. C. Beobachtungen über Kern-und Zelltheilung. *Ber. d. d. Bot. Gesellsch.*, Bd. II, Heft 7, p. 247.

1896. Wilson, E. B. The cell in development and inheritance. New York.

1899. Williams, C. L. The Origin of the Karyokinetic Spindle in Passiflora cœrulea Linn. *Proc. Calif. Acad. Sci.*, 3d Ser. (Bot.), Vol. I, p. 189.

EXPLANATION OF PLATE XXX.

All figures were drawn with the Abbé camera lucida. Objective, Zeiss Hom. Imm. $\frac{1}{12}$; Comp. ocular, No. 6.

Fig. 1. A young pollen-mother-cell. The cytoplasm consists of elongated meshes.

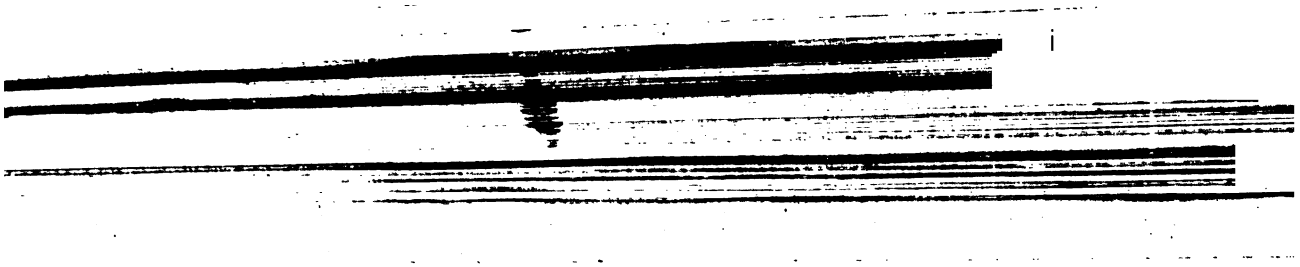
Fig. 2. Change in cytoplasm. The meshes of the layer immediately adjacent to the nucleus become smaller. Beginning of accumulation of granular matter around the nucleus.

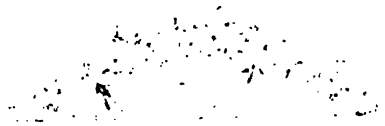
Fig. 3. Formation of violet-staining fibers in conical groups in the cytoplasm. The fibers parallel to the nuclear wall have become smooth.

Fig. 4. A dense granular zone has accumulated around the nucleus.

Fig. 5. Granular fibers have appeared in the granular zone.

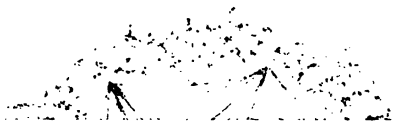
Fig. 6. Formation of cones. More or less parallel linin threads are seen in the nucleus.





EXPLANATION OF PLATE XXXI.

Fig. 7. The nuclear wall breaks down. The fibers are grouped in bundles.
Fig. 8. Nucleus with two especially prominent cones.
Figs. 9-10. Multipolar spindles; these occur but rarely.
Fig. 11. Indication of bipolar spindle.
Fig. 12. Perfect bipolar spindle.



EXPLANATION OF PLATE XXXII.

Fig. 13. Daughter chromosomes proceeding toward the poles.
Fig. 14. Daughter chromosomes at poles. Several small, red-staining bodies present in granular zone and cytoplasm.
Fig. 15. The daughter nuclei are surrounded by membranes.
Fig. 16. Felt-like zone around daughter nuclei.
Fig. 17. Multipolar diarchal spindle of daughter nuclei.
Fig. 18. Bipolar spindle of daughter nuclei.

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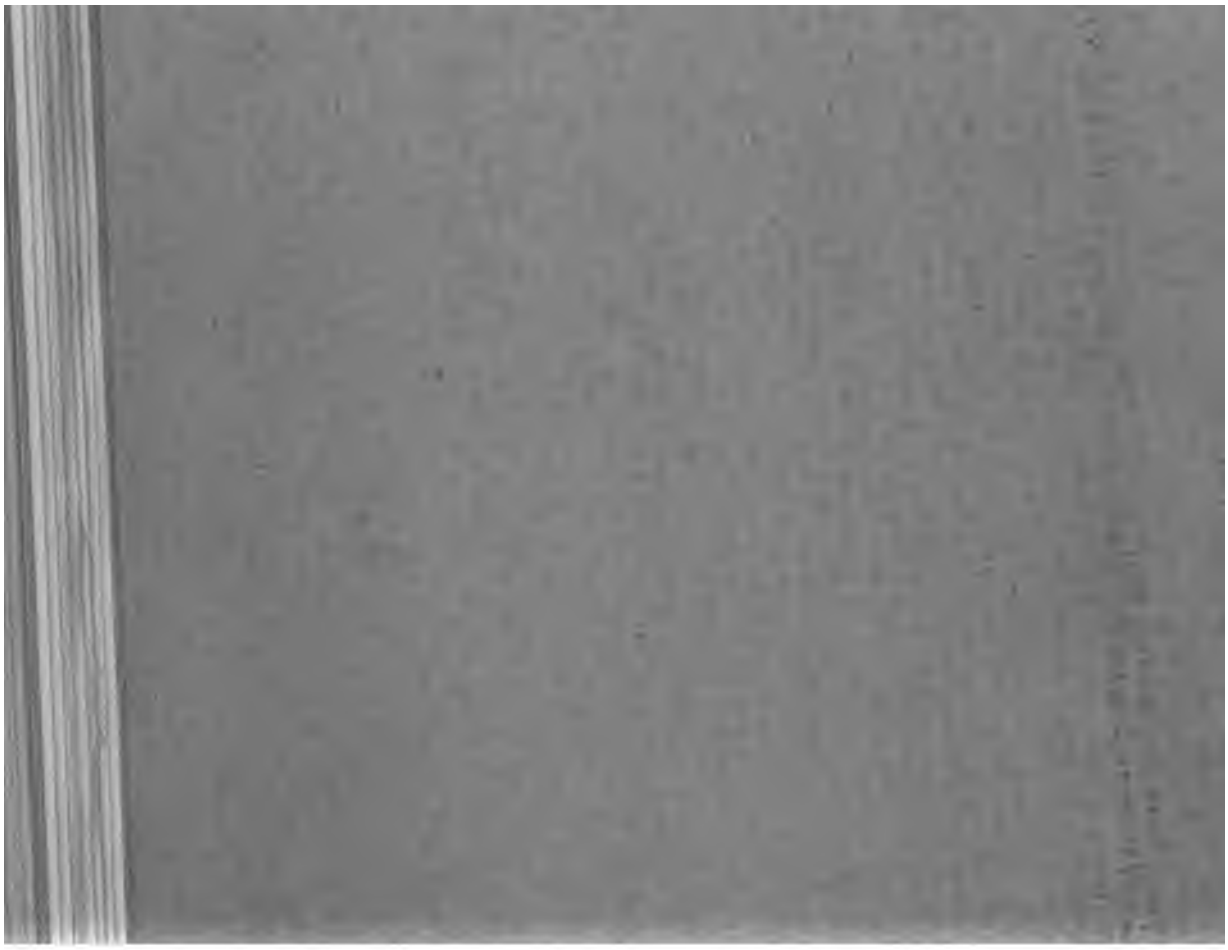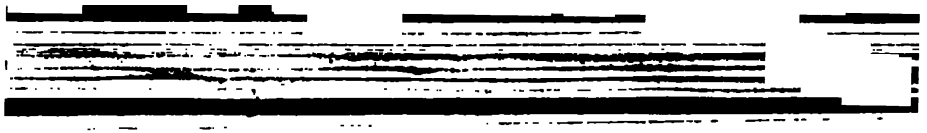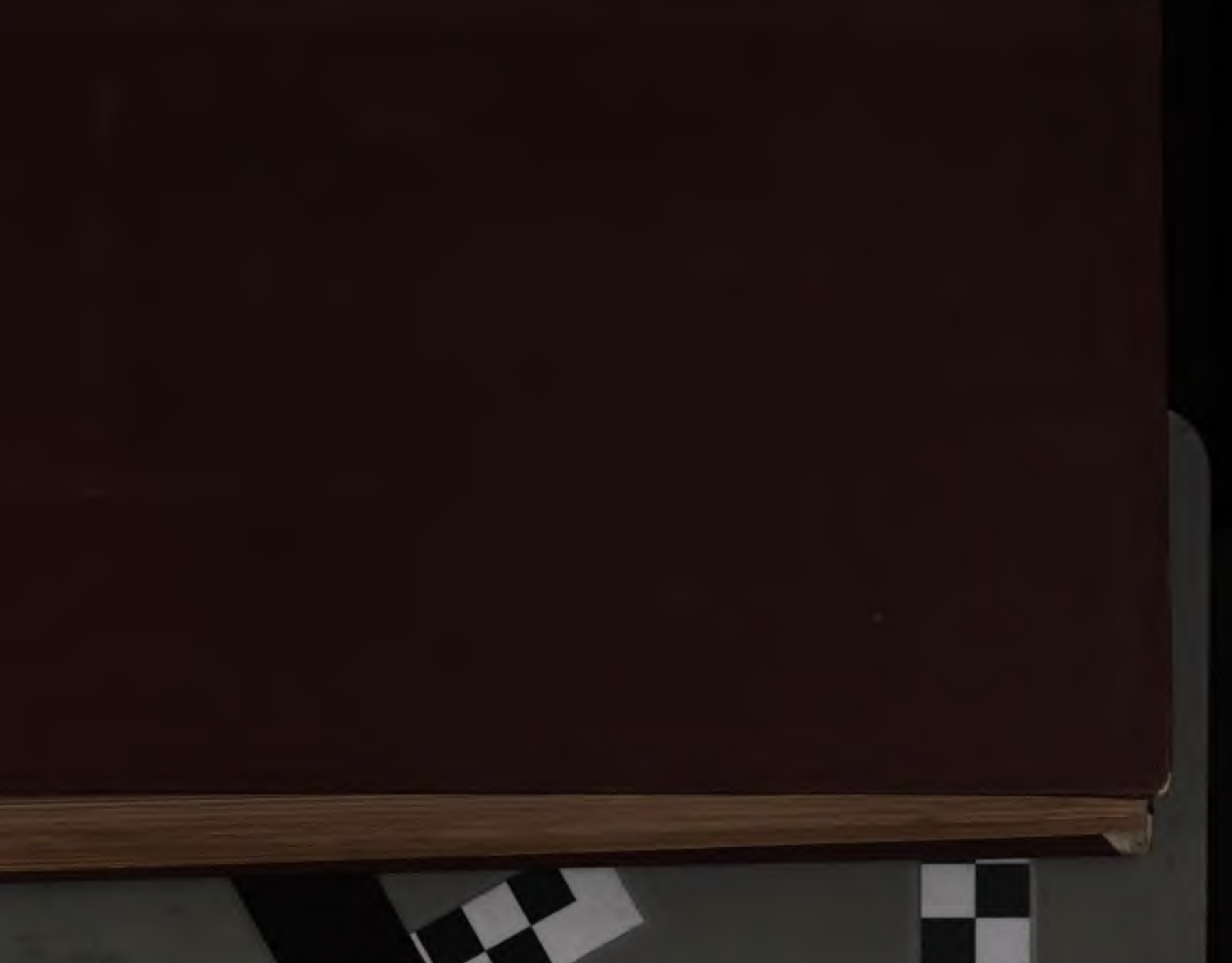
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